Appendix A

Sediment Data

Appendix A-1

Sediment Toxicity, Particle Size, and TOC

Appendix A-1: Sediment toxicity, particle size, and TOC.

Site Code	Site Name	SJR Sub-basin	Sample Date	Mean Survival (%)	Mean Survival of Control (%)	QA Code	Fine Sand (%)	Silt (%)	Clay (%)	Total Silt & Clay (%)	TOC (%)
SJC 509	Mt. House Creek at Mt. House Parkway	Delta	9/24/2002	86	92	NSG	35.32	30.78	31.5	62.28	0.89
SJC 515	Bear Creek at Lower Sacramento Road	Delta	5/29/2002	100	107	NSG	NS	NS	NS	NS	NS
SJC 515	Bear Creek at Lower Sacramento Road	Delta	9/18/2002	96	103	NSG	36.54	19.9	20.5	40.4	0.51
SJC 516	Unnamed Canal at Howard Road	DPR Delta	4/15/2003	86.25	100	NSG	32.28	43.94	21	64.94	1.82
SJC 516	Unnamed Canal at Howard Road	DPR Delta	3/1/2004	96	97	NSG	NS	NS	NS	NS	NS
SJC 516	Unnamed Canal at Howard Road	DPR Delta	6/21/2004	94	100	NSG	NS	NS	NS	NS	NS
SJC 516	Unnamed Canal at Howard Road	DPR Delta	3/7/2005	90	113	NSG	27.96	46.93	24.1	71.03	1.15
SJC 516	Unnamed Canal at Howard Road	DPR Delta	6/15/2005	90	92.3	NSG	25.01	50.8	20.67	71.47	1.25
SJC 516	Unnamed Canal at Howard Road	DPR Delta	9/19/2005	92.5	96.1	NSG	1.63	38.14	40.43	78.57	1.27
SJC 517	Mid Roberts Island Drain at Woodsbro Road	DPR Delta	4/15/2003	87.5	102	NSG	7.77	34.06	55	89.06	1.42
SJC 517	Mid Roberts Island Drain at Woodsbro Road	DPR Delta	3/1/2004	94	100	NSG	NS	NS	NS	NS	NS
SJC 517	Mid Roberts Island Drain at Woodsbro Road	DPR Delta	6/21/2004	93	99	NSG	NS	NS	NS	NS	NS
SJC 517	Mid Roberts Island Drain at Woodsbro Road	DPR Delta	3/7/2005	95	120	NSG	21.15	31.32	40.23	71.55	1.36
SJC 517	Mid Roberts Island Drain at Woodsbro Road	DPR Delta	6/15/2005	89	91	NSG	1.56	44.68	53.58	98.26	2.72
SJC 517	Mid Roberts Island Drain at Woodsbro Road	DPR Delta	9/19/2005	95	98.7	NSG	2.89	44.49	52.37	96.86	2.35
MER 007	Bear Creek near Bert Crane Road	Eastside	10/9/2001	93	97.4	NSG	83.72	6.63	5	11.63	0.21
MER 007	Bear Creek near Bert Crane Road	Eastside	5/28/2002	96	103	NSG	60.25	11.11	6	17.11	0.34
MER 007	Bear Creek near Bert Crane Road	Eastside	9/17/2002	78	82.7	NSG	57.69	10.1	9	19.1	0.29
MER 007	Bear Creek near Bert Crane Road	Eastside	4/8/2003	92.5	98.7	NSG	64.08	10.58	16	26.58	0.34
MER 546	Merced River at River Road	Eastside	5/28/2002	98	104	NSG	84.02	8.76	4.5	13.26	0.42
MER 579	Ingalsbe Slough at J17 Turlock Road	Eastside	5/28/2002	98	104	NSG	53.6	35.3	7.75	43.05	2.57
MER 579	Ingalsbe Slough at J17 Turlock Road	Eastside	9/17/2002	83	88	NSG	39.09	42.86	17	59.86	3.36
SJC 503	Lone Tree Creek at Austin Road	Eastside	10/9/2001	91	96.1	NSG	26.39	53.96	19	72.96	2.5
SJC 503	Lone Tree Creek at Austin Road	Eastside	5/29/2002	83	88	NSG	30.26	13.91	10.5	24.41	0.39
SJC 503	Lone Tree Creek at Austin Road	Eastside	9/18/2002	84	89.3	NSG	75.52	10.83	8	18.83	0.5
SJC 503	Lone Tree Creek at Austin Road	Eastside	4/9/2003	91.25	97.3	NSG	46.33	25.95	12	37.95	1.03
SJC 504	French Camp Slough at Airport Way	Eastside	5/29/2002	96	103	NSG	50.11	8	5.75	13.75	0.29
SJC 504	French Camp Slough at Airport Way	Eastside	9/18/2002	88	93.9	NSG	17.88	53.04	26	79.04	1.28
SJC 504	French Camp Slough at Airport Way	Eastside	4/9/2003	76.25	81.3	NSG	39.57	30.81	18	48.81	0.87

Site Code	Site Name	SJR Sub-basin	Sample Date	Mean Survival (%)	Mean Survival of Control (%)	QA Code	Fine Sand (%)	Silt (%)	Clay (%)	Total Silt & Clay (%)	TOC (%)
SJC 513	Calaveras River at Hwy 88	Eastside	9/18/2002	76	81.3	NSG	39.16	19.55	8	27.55	0.73
STC 501	TID 5 Harding Drain at Carpenter Road	Eastside	10/9/2001	83	86.8	NSG	63.77	22.65	6.5	29.15	0.31
STC 501	TID 5 Harding Drain at Carpenter Road	Eastside	4/8/2003	60	64	SL	57.49	23.97	14	37.97	0.68
MER 531	Salt Slough at Lander Avenue	Grassland	10/9/2001	89	93.4	NSG	64.45	18.08	14	32.08	0.55
MER 531	Salt Slough at Lander Avenue	Grassland	5/28/2002	93	98.7	NSG	*	*	*	*	*
MER 531	Salt Slough at Lander Avenue	Grassland	9/19/2002	91	97.3	NSG	29.08	46.21	24	70.21	0.73
MER 531	Salt Slough at Lander Avenue	Grassland	4/8/2003	90	96	NSG	31.17	45.17	23	68.17	0.83
MER 536	Mud Slough Upstream of SLD Terminus	Grassland	10/9/2001	93	97.4	NSG	67.49	22.49	9.5	31.99	0.56
MER 536	Mud Slough Upstream of SLD Terminus	Grassland	5/28/2002	96	103	NSG	47.59	33.62	18.5	52.12	0.6
MER 536	Mud Slough Upstream of SLD Terminus	Grassland	9/17/2002	93	98.7	NSG	64.61	21.35	12	33.35	0.42
MER 536	Mud Slough Upstream of SLD Terminus	Grassland	4/8/2003	77.5	82.7	NSG	53.84	27.39	17	44.39	0.42
MER 542	Mud Slough at San Luis Drain	Grassland	10/9/2001	88	92.1	NSG	80.24	9.15	9	18.15	0.41
MER 542	Mud Slough at San Luis Drain	Grassland	5/28/02	98	104	NSG	*	*	*	*	*
MER 542	Mud Slough at San Luis Drain	Grassland	9/17/2002	95	101	NSG	67.86	13.19	8.5	21.69	0.26
MER 542	Mud Slough at San Luis Drain	Grassland	4/8/2003	77.5	82.7	NSG	18.57	41.38	39	80.38	1.41
AMA 002	Sutter Creek at Hwy 49	Northeast	5/31/2002	98	104	NSG	32.58	4.84	4	8.84	0.54
CAL 003	N. Fork Calaveras River at Gold Strike Rd.	Northeast	5/31/2002	98	104	NSG	57.42	5.88	4.75	10.63	0.82
CAL 008	Calaveras River at Monte Vista Trailhead	Northeast	5/31/2002	78	82.7	NSG	26.85	3.24	3.75	6.99	0.4
ELD 004	Cosumnes River at Hwy 49	Northeast	5/31/2002	99	105	NSG	23.45	0.28	2.96	3.24	0.23
SAC 002	Mokelumne River at New Hope Road	Northeast	9/18/2002	89	94.7	NSG	79.11	4.39	4	8.39	0.18
SAC 002	Mokelumne River at New Hope Road	Northeast	4/9/2003	93.75	100	NSG	54.64	7.77	8	15.77	0.42
SAC 003	Cosumnes River at Michigan Bar Rd.	Northeast	10/9/2001	79	82.9	NSG	76.05	14.38	5	19.38	0.94
SAC 003	Cosumnes River at Michigan Bar Rd.	Northeast	5/31/2002	96	103	NSG	*	*	*	*	*
SJC 512	Mokelumne River at Van Assen Co. Park	Northeast	5/29/2002	85	90.7	NSG	24.03	27.39	39.75	67.14	4.3
STC 019	Orestimba Creek at River Road	Westside	10/9/2001	65	68.4	SL	32.08	38.28	23	61.28	0.63
STC 019	Orestimba Creek at River Road	Westside	5/29/2002	86	92	NSG	19.67	42.36	27	69.36	0.72
STC 019	Orestimba Creek at River Road	Westside	9/19/2002	41	44	SL	24.38	53.43	22	75.43	0.53
STC 019	Orestimba Creek at River Road	Westside	4/8/2003	55	58.7	SL	32.45	30.38	18	48.38	0.7
STC 030	Grayson Road Drain at Grayson	Westside	9/19/2002	58	61.3	SL	5.2	46.16	48.5	94.66	0.96

Site Code	Site Name	SJR Sub-basin	Sample Date	Mean Survival (%)	Mean Survival of Control (%)	QA Code	Fine Sand (%)	Silt (%)	Clay (%)	Total Silt & Clay (%)	TOC (%)
STC 030	Grayson Road Drain at Grayson	Westside	4/9/2003	0	0	SL	48.91	23.13	21	44.13	0.34
STC 030	Grayson Road Drain at Grayson	Westside	7/15/2003	17.8	20.32	SL	48.63	28.37	21	49.37	0.15
STC 030	Grayson Road Drain at Grayson	Westside	6/15/2005	38	39.2	SL	9.8	41.87	48.13	90	0.72
STC 040	Ingram Creek at River Road	Westside	9/24/2002	0	0	SL	6.79	41.8	51	92.8	0.83
STC 040	Ingram Creek at River Road	Westside	4/9/2003	18.75	20	SL	8.97	40.81	48	88.81	0.97
STC 040	Ingram Creek at River Road	Westside	7/15/2003	55.1	62.98	SL	14.06	44.63	40.5	85.13	0.58
STC 040	Ingram Creek at River Road	Westside	11/13/2003	2.5	3	SL	NS	NS	NS	NS	NS
STC 516	Del Puerto Creek at Vineyard Avenue	Westside	10/9/2001	0	0	SL	45.31	32.12	14.5	46.62	0.64
STC 516	Del Puerto Creek at Vineyard Avenue	Westside	5/29/2002	61	64.9	SL	24.08	47.59	25	72.59	0.91
STC 516	Del Puerto Creek at Vineyard Avenue	Westside	10/28/2002	5	5.79	SL	33.76	40.27	19	59.27	0.83
STC 516	Del Puerto Creek at Vineyard Avenue	Westside	4/9/2003	37.5	40	SL	24.93	51.17	21.5	72.67	0.88
STC 516	Del Puerto Creek at Vineyard Avenue	Westside	6/15/2005	39	40.3	SL	39.25	45.99	13.92	59.91	0.82
STC DP1	Del Puerto Creek 100 ft. upstream of Vineyard Avenue	Westside	10/28/2002	46	50.2	SL	19.59	30.03	44	74.03	1.77
STC 523	Del Puerto Creek at Hwy 33	Westside	10/28/2002	79	111	NSG	39.56	43.99	15	58.99	0.57
STC 524	Del Puerto Creek at Rogers	Westside	10/28/2002	33	46.4	SL	26.87	22.17	33	55.17	2.3

*	Broken Sample Container
#	Not calculated
TIE	TIE completed in addition to Sediment Toxicity
NS	Not submitted
ND	Not detected
<RL*	Detected at concentrations below the reporting limit

QA Codes

SL	Significant compared to negative control based on statistical test, alpha of less than 5%, AND less than evaluation threshold (Both criteria met)
NSG	Not significant compared to negative control based on statistical test, alpha of 5%, and is above the evaluation threshold (No criteria met)

Appendix A-2

Sediment Pyrethroid Pesticide Results

Appendix A-2: Sediment pyrethroid pesticide results.

Pyrethroid results for Del Puerto Creek Upstream Study

Site Code		STC 516	STC DP1	STC 523	STC 524
Site Name		DPC @ Vineyard	DPC 100 ft. upstream	DPC @ 33	DPC @ Rogers
Sample Date		10/28/2002	10/28/2002	10/28/2002	10/28/2002
Pyrethroids	**Reporting limit ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**
Bifenthrin	5.0	**7.51**	<RL	ND	ND
Lambda-cyhalothrin	5.0	ND	ND	ND	ND
Permethrin	5.0	ND	ND	ND	ND
Cyfluthrin	5.0	ND	<RL	ND	ND
Esfenvalerate	5.0	ND	ND	ND	ND
Fenvalerate	5.0	ND	ND	ND	ND

<RL = Detected at less than the reporting limit.
ND = Not detected.

Appendix A-2: Sediment pyrethroid pesticides results.

Pyrethroid results for June 2005 and September 2005								
Site Code		SJC 504	SJC 531	STC 516	SJC 504	SJC 531	SJC 516	SJC 517
Site Name		French Camp Slough	Lone Tree @ E.B.	DPC @ Vineyard	French Camp Slough	Lone Tree @ E.B.	Unkown Supply @ Howard	Mid Robers Island
Sample Date		6/15/2005	6/15/2005	6/15/2005	9/19/2005	9/19/2005	9/19/2005	9/19/2005
Pyrethroids	**Reporting limit ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**
Bifenthrin	1.00	**1.02**	ND	ND	**2.44**	**1.64**	ND	ND
Cyfluthrin - 1	3.00	ND	ND	ND	ND	ND	ND	ND
Cyfluthrin - 2	3.00	ND	ND	ND	ND	ND	ND	ND
Cyfluthrin - 3	3.00	ND	ND	ND	ND	ND	ND	ND
Cyfluthrin - 4	3.00	ND	ND	ND	ND	ND	ND	ND
Cypermethrin - 1	3.00	ND	ND	**1.85***	ND	ND	ND	ND
Cypermethrin - 2	3.00	ND	ND	ND	ND	ND	ND	ND
Cypermethrin - 3	3.00	ND	ND	**2.03***	ND	ND	ND	ND
Cypermethrin - 4	3.00	ND	ND	ND	ND	ND	ND	ND
Deltamethrin	1.00	#	#	#	ND	ND	ND	ND
Es-fenvalerate - 1	2.00	ND	ND	ND	ND	ND	ND	ND
Es-fenvalerate - 2	2.00	ND	ND	ND	ND	ND	ND	ND
Lamda-cyhalothrin - 1	1.00	ND	ND	ND	ND	ND	ND	ND
Lamda-cyhalothrin - 2	1.00	ND	ND	ND	**2.19**	ND	ND	ND
Permethrin - 1	4.00	ND	ND	ND	ND	ND	ND	ND
Permethrin - 2	4.00	ND	ND	ND	ND	ND	ND	ND

* = Result reported is less than reporting limit.
ND = Not detected.
# = Not analyzed

Appendix A-3

Organophosphate Pesticides Results

Appendix A-3: Organophosphate pesticides results.

Site Code	Site Name	Sample Date	Chlorpyrifos (ng/g)*	Diazinon (ng/g)*
MER007	Bear Creek near Bert Crane Rd.	5/28/2002	ND	ND
MER531	Salt Slough at Lander Ave.	5/28/2002	ND	ND
MER536	Mud Slough Upstream of SLD Terminus	5/28/2002	ND	ND
MER542	Mud Slough at San Luis Drain	5/28/2002	ND	ND
MER546	Merced River at River Rd.	5/28/2002	ND	ND
MER579	Ingalsbe Slough at J17 Turlock Rd.	5/28/2002	ND	ND
SJC503	Lone Tree Creek at Austin Rd.	5/29/2002	ND	ND
SJC504	French Camp Slough at Airport Way	5/29/2002	ND	ND
SJC515	Bear Creek at Lower Sacramento Rd.	5/29/2002	ND	ND
SJC512	Mokelumne River at Van Assen Co. Park	5/29/2002	ND	ND
STC019	Orestimba Creek at River Road	5/29/2002	ND	ND
STC516	Del Puerto Creek at River Rd.	5/29/2002	5.6	4.7
AMA002	Sutter Creek at Hwy 49	5/31/2002	ND	ND
CAL003	N. Fork Calaveras River at Gold Strike Rd.	5/31/2002	ND	ND
CAL008	Calaveras River at Monte Vista Trailhead	5/31/2002	ND	ND
ELD004	Cosumnes River at Hwy 49	5/31/2002	ND	ND
SAC003	Cosumnes River at Michigan Bar Rd.	5/31/2002	ND	ND
MER007	Bear Creek near Bert Crane Rd.	9/17/2002	ND	ND
MER536	Mud Slough Upstream of SLD Terminus	9/17/2002	ND	ND
MER542	Mud Slough at San Luis Drain	9/17/2002	ND	ND
MER579	Ingalsbe Slough at J17 Turlock Rd.	9/17/2002	ND	ND
SAC002	Mokelumne River at New Hope Rd.	9/18/2002	ND	ND
SJC503	Lone Tree Creek at Austin Rd.	9/18/2002	ND	ND
SJC504	French Camp Slough at Airport Way	9/18/2002	ND	ND
SJC515	Bear Creek at Lower Sacramento Rd.	9/18/2002	ND	ND
SJC513	Calaveras River at Hwy 88	9/18/2002	ND	ND
MER531	Salt Slough at Lander Ave.	9/19/2002	ND	ND
STC019	Orestimba Creek at River Road	9/19/2002	ND	3.7
STC030	Grayson Road Drain at Grayson Rd.	9/19/2002	ND	ND
SJC509	Mt. House Creek at Mt. House Parkway	9/24/2002	ND	ND
STC040	Ingram Creek at Vineyard Ave.	9/24/2002	ND	ND

* Analysis using ELISA.

Appendix A-3: Organophosphate pesticides results.

Site Code		STC 516	STC DP1	STC 523	STC 524
Site Name		DPC @ Vineyard	DPC 100 ft. upstream	DPC @ 33	DPC @ Rogers
Sample Date		10/28/2002	10/28/2002	10/28/2002	10/28/2002
Organophosphate Pesticides	**Reporting limit ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**
Aspon	10.0	ND	ND	ND	ND
Azinphos-ethyl	10.0	ND	ND	ND	ND
Carbophenothion	10.0	ND	ND	ND	ND
Chlorfenvinphos	10.0	ND	ND	ND	ND
Chlorpyrifos	10.0	ND	ND	ND	ND
Chlorpyrifos methyl	10.0	ND	ND	ND	ND
Ciodrin(Crotoxyphos)	10.0	ND	ND	ND	ND
Coumaphos	10.0	ND	ND	ND	ND
Demeton-S	10.0	ND	ND	ND	ND
Diazinon	5.00	ND	ND	ND	ND
Dibrom(naled)	10.0	ND	ND	ND	ND
Dichlofenthion	10.0	ND	ND	ND	ND
Dichlorvos	10.0	ND	ND	ND	ND
Dicrotophos	10.0	ND	ND	ND	ND
Dimethoat	10.0	ND	ND	ND	ND
Dioxathion	10.0	ND	ND	ND	ND
Disulfoton	10.0	ND	ND	ND	ND
Ethion	10.0	ND	ND	ND	ND
Famphur	10.0	ND	ND	ND	ND
Fenchlorophos(Ronnel)	10.0	ND	ND	ND	ND
Fenitrothion	10.0	ND	ND	ND	ND
Fensulfothion	10.0	ND	ND	ND	ND
Fenthion(Mercaptophos)	10.0	ND	ND	ND	ND
Fonofos (Dyfonate)	10.0	ND	ND	ND	ND
Guthion(Azinphos methyl)	10.0	ND	ND	ND	ND
Leptophos	10.0	ND	ND	ND	ND
Methidathion	10.0	ND	ND	ND	ND
Methyl parathion	10.0	ND	ND	ND	ND
Parathion	10.0	ND	ND	ND	ND
Phorate	10.0	ND	ND	ND	ND
Phosdrin(Mevinphos)	10.0	ND	ND	ND	ND
Phosmet	10.0	ND	ND	ND	ND
Phosphamidon	10.0	ND	ND	ND	ND
Prophos(Ethoprop)	10.0	ND	ND	ND	ND
Sulprofos(Bolstar)	10.0	ND	ND	ND	ND

Site Code		STC 516	STC DP1	STC 523	STC 524
Site Name		DPC @ Vineyard	DPC 100 ft. upstream	DPC @ 33	DPC @ Rogers
Sample Date		10/28/2002	10/28/2002	10/28/2002	10/28/2002
Organophosphate Pesticides	**Reporting limit ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**
Terbufos	10.0	ND	ND	ND	ND
Tetrachlorvinphos(stirifos)	10.0	ND	ND	ND	ND
Thionzin(Thionazin)	10.0	ND	ND	ND	ND
Tokuthion	10.0	ND	ND	ND	ND
Tributylphosphorotrithioite(Merphos)	10.0	ND	ND	ND	ND
Trichlorfon	10.0	ND	ND	ND	ND
Trichloronate	10.0	ND	ND	ND	ND

Appendix A-3: Organophosphate Pesticides

Site Code		STC501	STC019	STC030	STC040	STC516
Site Name		TID Lateral 5	Orestimba @ RR	Grayson Drain	Ingram @ RR	DPC @ Vineyard
Sample Date		4/8/2003	4/8/2003	4/9/2003	4/9/2003	4/9/2003
Organophosphate Pesticides	**Reporting limit ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**	**ppb (ng/g)**
Chlorpyrifos	2.00	**1.82**	ND	**6.18**	**1.67**	**9.20**
Coumaphos	5.00	ND	ND	ND	ND	ND
Demeton-S	5.00	ND	ND	ND	ND	ND
Diazinon	1.00	**2.26**	ND	**1.26**	**1.23**	**1.23**
Dibrom(naled)	5.00	ND	ND	ND	ND	ND
Dichlorvos	5.00	ND	ND	ND	ND	ND
Disulfoton	5.00	ND	ND	ND	ND	ND
Fenchlorophos(Ronnel)	5.00	ND	ND	ND	ND	ND
Fensulfothion	5.00	ND	ND	ND	ND	ND
Fenthion(Mercaptophos)	5.00	ND	ND	ND	ND	ND
Guthion(Azinphos methyl)	5.00	ND	ND	ND	ND	ND
Methyl parathion	5.00	ND	ND	ND	ND	ND
Phorate	5.00	ND	ND	ND	ND	ND
Phosdrin(Mevinphos)	5.00	ND	ND	ND	ND	ND
Prophos(Ethoprop)	5.00	ND	ND	ND	ND	ND
Sulprofos(Bolstar)	5.00	ND	ND	ND	ND	ND
Tetrachlorvinphos(stirifos)	5.00	ND	ND	ND	ND	ND
Tokuthion	5.00	ND	ND	ND	ND	ND
Tributylphosphorotrithioite(Merphos)	5.00	ND	ND	ND	ND	ND
Trichloronate	5.00	ND	ND	ND	ND	ND

Appendix B

Toxicity Identification Evaluations

Conducted by the University of California Davis Department of Environmental Toxicology Marine Pollution Studies Laboratory

Appendix B-1

Del Puerto Creek at Vineyard Road May 29, 2002 and September 11, 2002 sampling events

Toxicity Identification Evaluation Results

Region 5 - Station 541STC516

Surface Water Ambient Monitoring Program

By the University of California, Davis - Department of Environmental Toxicology

Marine Pollution Studies Laboratory
34500 Coast Route One, Granite Canyon
Monterey, CA 93940

Submitted to the Central Valley Regional Water Quality Control Board – Sacramento Region 5

December 2002

Introduction

A sediment sample collected from Region 5 as part of the Surface Water Ambient Monitoring Program (SWAMP) was tested for toxicity to *Hyalella azteca* using established testing protocols. Because the sample was significantly toxic to the test organism, a toxicity identification evaluation (TIE) was conducted. TIEs are designed to proceed in three phases. The purpose of a Phase 1 TIE is to characterized the cause of toxicity. Information from the Phase 1 characterization may then be used in subsequent Phase 2 (identification) and Phase 3 (confirmation) TIEs. Based on the results of initial toxicity tests, two Phase 1 TIEs were conducted to investigate the causes of toxicity. This report presents the data obtained from these TIEs, including the mean percent survival of amphipods after exposure to various TIE treatments, water quality measurements of test solutions, and copies of the original data sheets and quality assurance forms.

Methods

Sample Handling

Sediment samples were collected on May 29 and September 11, 2002, under the supervision of Jay Rowan (Central Valley Regional Water Quality Control Board). Samples were transported on ice and in the dark to the Marine Pollution Studies Laboratory at Granite Canyon for initial toxicity testing, which began within 14 days of collection (see attached copy of chain of custody form). Pore water was extracted from the first sample on June 6, and an initial test initiated on June 7. After the termination of the initial test, the first TIE was initiated on June 14. Pore water was extracted from the second sample on September 12, and initial tests with sediment and pore water were initiated on September 13. The pore water TIE was initiated on September 16.

TIE Methods

The following Phase 1 TIE treatments were performed on a dilution series of each sample (US EPA 1991). Sample concentrations in the first TIE were 0 (treatment blank), 10, 25, 50, and 100%. Concentrations in the second TIE were 0 (blank), 1, 5, 10, and 50%. The treatment blank was control water that underwent the same manipulation as the sample.

Treatments:

• Baseline - Toxicity test on un-manipulated sample. Concentrations were chosen to bracket the effect concentration of the sample and might differ from initial test.

• Centrifugation - Used to determine whether toxicants are associated with particles. Also used as a pretreatment step for the column treatments. Because TIEs were conducted on pore water that was extracted via centrifugation, this treatment served as the Baseline.

• Aeration - Samples are aerated to determine if their toxicity is due to volatile compounds or surfactants.

• EDTA (Disodium Ethylenediaminetetraacetic acid) - EDTA is an organic chelating agent that preferentially binds with divalent metals, such as copper, nickel, lead, zinc, cadmium, mercury, and other transition metals to form non-toxic complexes. It will not complex with anionic forms of metals such as selenids, chromates and hydrochromates.

• STS (Sodium Thiosulfate) - Addition of STS, a reducing agent, to a sample containing oxidants (chlorine, bromine), results in a reduction reaction that may decrease sample toxicity. STS is also a chelating agent for some cationic metals.

• pH Shift - Changes in pH can affect solubility, polarity, volatility, stability and speciation of a compound, thereby affecting its bioavailability and toxicity. Shifting pH is designed to determine how much sample toxicity can be attributed to volatile, sublateable or oxidizeable compounds. Shifts in pH can also be combined with Aeration or solid-phase extraction with the C8 Column.

• C8 Column - The C8 Column is designed to remove non-polar organic compounds from the sample. In the manipulation, reverse phase liquid chromatography is applied to extract nonionic organic toxicants from the aqueous sample. Column can be eluted with methanol and resulting eluate tested to determine if substances removed by the column are indeed toxic.

• Oasis Column – Another type of solid-phase extraction column designed to remove non-polar organic compounds from the sample. This column has been shown to remove pyrethroid pesticides.

• PBO (Piperonyl Butoxide) - PBO is a metabolic inhibitor that removes the toxicity associated with metabolically activated pesticides such as diazinon and chlorpyrifos. An increase of toxicity with the PBO treatments can indicate the presence of non-metabolically activated compounds such as pyrethroid pesticides.

In addition to the standard Phase 1 treatments, two experimental treatments were used to characterize pyrethroid toxicity. The temperature treatment mimics the Baseline treatment but the organisms are exposed at 15°C instead of 23°C. Type 1 pyrethroid pesticides are known to be more toxic at colder temperatures. Because Type 1 compounds are among the more commonly used pyrethroids, toxicity at colder temperatures may help characterize this class of pesticides as the cause of toxicity. The Enzyme treatment uses a pyrethroid-binding enzyme to reduce the bioavailability of the pesticides.

Exposures were conducted in 20 mL glass scintillation vials (5 to 10 replicates) containing 15 mL treated sample and one amphipod. Acute exposures were conducted for 96 hours, following US EPA 1993.

Physical and Chemical Measurements

Water quality parameters of dissolved oxygen, pH and conductivity were measured using a Hach SensION© selective ion meter with appropriate electrodes; and ammonia was measured using a Hach 2010 spectrophotometer. Temperature was measured using a continuously recording thermograph and thermometer. Concentrations of the organophosphate pesticides chlorpyrifos and diazinon were measured using enzyme-linked immunosorbent assays (ELISA, Strategic Diagnostics Inc, Newark, DE). The California Department of Fish and Game Nimbus Laboratory conducted pesticide analyses under the supervision of Dave Crane. Pyrethroids (EPA Method 1660), organochlorines (EPA Method 8081), and organophosphates (EPA Method 8141A) were analyzed in sediment and porewater

Data Interpretation

Treatment blanks were evaluated to determine if sample manipulations added toxic artifacts. Treatment data were then compared to one another using the toxic unit approach. Toxic units (TU) were calculated by dividing 100 by the LC50 calculated from each treatment dilution series. More toxic units indicate a more toxic sample.

Results and Discussion

Hyalella TIE 6/14/02

The initial test was significantly toxic to *Hyalella* in a 96-hour acute exposure (Table 1). Toxic units were not calculated as part of the initial test because there was no dilution series.

Several Phase 1 treatments reduced toxicity. All of the pH treatments reduced toxicity to some extent, but the pH 3 Shift reduced toxicity the most (7.9 TU to 3.5 TU). Shifting pH can reduce toxicity by converting ionic compounds to more volatile or precipitant forms, and can affect the ionization state of polar toxicants, thus making them more or less volatile (US EPA 1988). It is not clear what compounds were being affected by the pH shifts. The Column treatments also reduced toxicity, but the pH 3 Column was the most successful. Because the pH 3 Column reduced toxicity to the same level as the pH 3 Shift alone, this reduction could be attributed to the pH shift. Some toxicity was added back in both Column Eluates. PBO increased toxicity by a factor of 2.5, suggesting the presence of pyrethroid pesticides.

Organophosphate pesticides were below the threshold values for *Hyalella*, and water quality parameters were all within the tolerance limits of the organism (Unionized Ammonia LC50 = 4.7 mg/L, Table 2).

Table 1. Mean percent survival of *Hyalella*, concentrations of organophosphate pesticides, and toxic units from Phase 1 TIE treatments conducted on 6/14/02.

Treatment	Toxic Units	Percent Sample 0%	10%	25%	50%	100%	Chlorpyrifos (μg/L)	Diazinon (μg/L)
Initial Test	NA	100				0	0.056	0.047
Baseline	7.9	100	60	20	0	0	NA	NA
EDTA	6.3	100	80	20	0	0	NA	NA
STS	11.5	100	40	20	0	0	NA	NA
pH 3 Shift	3.5	100	80	60	0	0	NA	NA
pH 11 Shift	5.0	100	80	40	0	20	NA	NA
pH 3 Aeration	5.1	100	80	40	0	0	NA	NA
pH Ambient Aeration	5.4	100	100	20	0	0	NA	NA
pH 11 Aeration	4.1	100	100	40	20	20	NA	NA
pH 3 C8 Column	3.7	100	80	60	20	20	NA	NA
pH 3 C8 Eluate	1.4	100	80	100	100	0	NA	NA
pH Ambient C8 Column	4.8	100	80	40	20	0	NA	NA
pH Ambient C8 Eluate	1.9	100	80	100	60	0	NA	NA
Oasis Column	4.6	100	100	40	0	0	NA	NA
PBO	20.0	100	0	0	0	0	NA	NA

Table 2. Water quality measurements for Phase 1 TIE treatments conducted on 6/14/02.

Treatment	Water Quality Parameter pH	Dissolved Oxygen (mg/L)	Conductivity (μS/cm)	Total Ammonia (mg/L)	Un-ionized Ammonia (mg/L)
Baseline	7.64	3.11	1657	3.6	0.076
EDTA	7.61	3.99	1666	3.3	0.065
STS	7.77	2.42	1740	3.4	0.096
pH 3 Shift	7.56	8.37	2590	3.8	0.067
pH 11 Shift	7.71	8.05	2980	3.1	0.076
pH 3 Aeration	7.74	8.55	2260	4.0	0.106
pH Ambient Aeration	8.52	5.50	1662	3.3	0.463
pH 11 Aeration	7.68	8.40	2920	3.0	0.069
pH 3 C8 Column	7.61	8.46	2540	4.0	0.079
pH 3 C8 Eluate	8.14	7.43	700	ND	ND
pH Ambient C8 Column	7.90	5.82	1691	2.7	0.102
pH Ambient C8 Eluate	8.18	7.50	690	0.2	0.006
Oasis Column	8.12	6.80	1915	3.4	0.092
PBO	7.84	3.10	1683	3.3	0.047

Hyalella TIE 9/16/02

The initial test was significantly toxic to *Hyalella* in a second 96-hour acute exposure (Table 3). Toxic units were calculated as part of the initial test because there was a dilution series. *Hyalella* exposed in sediment also demonstrated complete mortality. The pH 3 Shift slightly reduced toxicity in the second Phase 1 TIE, but no other treatments were successful. The Baseline 15°C and PBO treatments both increased toxicity, again suggesting the presence of pyrethroids. The Pyrethroid Enzyme mitigated toxicity up to 48 hours (data not shown), but because the enzyme breaks down, 96-hour toxicity was not reduced. Future use of this experimental treatment will include daily renewals.

Pesticide concentrations in pore water and sediment are summarized in Table 4. Concentrations of the organochlorines are below threshold values for *Hyalella*, and organophosphates were not detected. The concentration of the pyrethroid bifenthrin might be affecting the test organisms. We were unable to find published *Hyalella* LC50 values for bifenthrin, but the pore water concentration in this sample was similar to published LC50 values for a variety of other aquatic bioassay organisms (Table 5).

Water quality parameters were all within the tolerance limits of the organism (Table 6). The ammonia concentration in the Enzyme treatment was elevated because the enzyme is prepared with ammonium sulfate. The concentration of unionized ammonia was still below the *Hyalella* threshold (Unionized Ammonia LC50 = 4.7 mg/L).

Table 3. Mean percent survival of *Hyalella*, concentrations of organophosphate pesticides, and toxic units from Phase 1 TIE treatments conducted on 9/16/02.

Treatment	Toxic Units	Percent Sample 0%	10%	25%	50%	100%	Chlorpyrifos (µg/L)	Diazinon (µg/L)
Initial Test	20.0	100	0	0	0	0	ND	0.039
		0%	1%	5%	10%	50%		
Baseline 23°C	17.9	100	100	60	20	0	NA	NA
Baseline 15°C	200.0	100	0	0	0	0	NA	NA
pH 3 Shift	13.9	100	100	100	0	10	NA	NA
pH 11 Shift	15.9	100	100	60	30	0	NA	NA
pH Ambient C8 Column	45.5	80	80	0	0	0	NA	NA
pH Ambient C8 Eluate	12.8	100	90	90	20	0	NA	NA
PBO	76.9	100	70	0	0	0	NA	NA
Enzyme	25.0	100	100	40	10	0	NA	NA

Table 4. Select pesticide concentrations from sediment and pore water tested on 9/16/02.

	Pore Water Concentration (μg/L)	Method Blank (μg/L)	Laboratory Spike (% Recovery)	Sediment Concentration (ng/g dry wt.)	Method Blank (μg/L)	Laboratory Spike (% Recovery)
Pyrethroids						
Bifenthrin	0.048	ND	93.5	43.2	ND	93.5
Permethrin	ND	ND	NA	20.4	ND	NA
Organochlorines						
DDE, o,p’	0.003	ND	94.5	1.37	ND	94.0
DDE, p,p’	0.056	ND	100	39.5	ND	97.0
DDT, o,p’	ND	ND	99.0	1.28	ND	106
DDT, p,p’	0.014	ND	100	14.7	ND	104
Dieldrin	0.004	ND	95.0	1.54	ND	102
Endosulfan I	0.003	ND	100	0.78	ND	85.6
Endosulfan sulfate	ND	ND	105	7.63	ND	90.0
HCH, alpha	0.004	ND	100	ND	ND	93.2
Mirex	0.056	ND	94.1	0.56	ND	91.3
Oxadiazone	0.023	ND	93.3	ND	ND	93.3

Table 5. Bifenthrin LC50 values for various aquatic organisms.

Organism	Test Duration	LC50 (μg/L)	Reference
Ceriodaphnia dubia	48-hour	0.078	CDFG – ATL Report P2161-2 (1999)
Daphnia magna	48-hour	0.320	Mokry and Hoagland (1990)
Pimephales promelas	96-hour	0.780	CDFG – ATL Report P2161-1 (1999)
Americamysis bahia	96-hour	0.004	Office of Pesticide Programs (2000)
Onchorhynchus mykiss	96-hour	0.150	Office of Pesticide Programs (2000)

Table 6. Water quality measurements for Phase 1 TIE treatments conducted on 9/16/02.

	Water Quality Parameter				
Treatment	pH	Dissolved Oxygen (mg/L)	Conductivity (μS/cm)	Total Ammonia (mg/L)	Un-ionized Ammonia (mg/L)
Baseline 23°C	8.07	8.16	1148	0.6	0.033
Baseline 25°C	8.07	8.16	1148	0.6	0.033
pH 3 Shift	8.11	8.06	1559	1.0	0.060
pH 11 Shift	8.24	8.12	1485	1.1	0.087
pH Ambient C8 Column	8.15	8.44	1127	0.7	0.046
pH Ambient C8 Eluate	8.35	8.50	640	ND	ND
PBO	8.08	7.93	1102	0.8	0.045
Enzyme	7.94	8.08	1221	23.5	0.968

Conclusion

Results from both sediment pore water TIEs were consistent. Toxicity was increased by adding PBO and by decreasing test temperature. Both lines of evidence suggest pyrethroid pesticides. Chemical analysis indicated sample concentrations of bifenthrin above toxicity thresholds for mysids. Bifenthrin toxicity to *Hyalella* is unknown.

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Appendix B-2

Ingram Creek at River Road
November 13, 2003 sampling event

Toxicity Identification Evaluation Results

Region 5 - Station 541STC040

Surface Water Ambient Monitoring Program

By the University of California, Davis - Department of Environmental Toxicology

Marine Pollution Studies Laboratory
34500 Coast Route One, Granite Canyon
Monterey, CA 93940

Submitted to the Central Valley Regional Water Quality Control Board – Sacramento Region 5

December 2003

Introduction

A sediment sample collected from Region 5 as part of the Surface Water Ambient Monitoring Program (SWAMP) was tested for toxicity to *Hyalella azteca* using established testing protocols. Because the sample was significantly toxic to the test organism, a toxicity identification evaluation (TIE) was conducted. TIEs are designed to proceed in three phases. The purpose of a Phase 1 TIE is to characterized the cause of toxicity. Information from the Phase 1 characterization may then be used in subsequent Phase 2 (identification) and Phase 3 (confirmation) TIEs. Based on the results of initial toxicity tests, two Phase 1 TIEs were conducted to investigate the causes of toxicity. This report presents the data obtained from these TIEs, including the mean percent survival of amphipods after exposure to various TIE treatments, water quality measurements of test solutions, and copies of the original data sheets and quality assurance forms.

Methods

Sample Handling

A sediment sample was collected on November 13, 2003, under supervision of Jay Rowan (Central Valley Regional Water Quality Control Board). The sample was transported on ice in the dark to the Marine Pollution Studies Laboratory at Granite Canyon for initial toxicity testing, which began within 14 days of collection (see original chain of custody form). Pore water was extracted from the sample on November 19, and an initial test started on November 20, 2003. After termination of the initial test, the first TIE was initiated on December 4. A solid-phase test was conducted simultaneously using standard *H. Azteca* 10-day protocol (US EPA 2000).

TIE Methods

The following Phase 1 TIE treatments were performed on a dilution series of each sample (US EPA 1991). Sample concentrations in the initial test were 0 (treatment blank), 10, 25, 50, and 100%. Concentrations in the second TIE were 0 (blank), 50, and 100%. The treatment blank was control water that underwent the same manipulation as the sample.

Treatments:

• Baseline - Toxicity test on un-manipulated sample. Concentrations were chosen to bracket the effect concentration of the sample and might differ from initial test.

• Centrifugation - Used to determine whether toxicants are associated with particles. Also used as a pretreatment step for the column treatments. Because TIEs were conducted on pore water that was extracted via centrifugation, this treatment served as the Baseline.

• C18 Column – The C8 column is designed to remove non-polar organic compounds from the sample. In the manipulation, reverse phase liquid chromatography is applied to extract nonionic organic toxicants from the aqueous sample. Column can be eluted with methanol and resulting eluate tested to determine if substances removed by the column are indeed toxic.

• PBO (Piperonyl Butoxide) - PBO is a metabolic inhibitor that removes the toxicity associated with metabolically activated pesticides such as diazinon and chlorpyrifos. An increase of toxicity with the PBO treatments can indicate the presence of non-metabolically activated compounds such as pyrethroid pesticides.

In addition to the standard Phase 1 treatments, two experimental treatments were used to characterize pyrethroid toxicity. The temperature treatment mimics the Baseline treatment but the organisms are exposed at 15°C instead of 23°C. Type 1 pyrethroid pesticides are known to be more toxic at colder temperatures. Because Type 1 compounds are among the more commonly used pyrethroids, toxicity at colder temperatures may help characterize this class of pesticides as the cause of toxicity. The Enzyme treatment uses a carboxylesterase enzyme to break down pyrethroids and reduce the bioavailability of the pesticides. A combination treatment consisting of the enzyme and PBO was also used.

Exposures were conducted in 20 mL glass scintillation vials (5 to 10 replicates) containing 15 mL treated sample and five amphipod. Acute exposures were conducted for 96 hours, following US EPA 1993.

Physical and Chemical Measurements

Water quality parameters of dissolved oxygen, pH and conductivity were measured using a Hach SensION© selective ion meter with appropriate electrodes; and ammonia was measured using a Hach 2010 spectrophotometer. Temperature was measured using a continuously recording thermograph and thermometer. Concentrations of the organophosphate pesticides chlorpyrifos and diazinon were measured using enzyme-linked immunosorbent assays (ELISA, Strategic Diagnostics Inc, Newark, DE).

Data Interpretation

Treatment blanks were evaluated to determine if sample manipulations added toxic artifacts. Treatment data were then compared to one another using based on organism response.

Results and Discussion

Hyalella TIE 6/14/02

The initial test was significantly toxic to *H. Azteca* at the 50% concentration in a 96-hour acute exposure (Table 1). Concentrations below 50% were not used in the TIE because they were not toxic to *H. Azteca*. There was no toxicity observed in the treatment blanks. At 48 hours the C18 Column had successfully reduced toxicity and there is evidence of add-back in the C18 eluate treatment. This result suggests an organic contaminant is a potential cause of toxicity. The enzyme treatment was also somewhat successful at reducing toxicity at 48 hours, indicating a pyrethroid might be contributing to the toxicity. By 96 hours the reductions in toxicity were encompassed by the toxic signal (Table 1). The organophosphate pesticides chlorpyrifos and diazinon were not detected in the sample, and water quality parameters were all within tolerance limits of the organism (Table 2).

Because this station is influenced by agricultural inputs, it is likely the toxicity was cause by organic contaminants. Partial reduction of toxicity by the C18 Column and partial add-back in the C18 Eluate support this conclusion.

References

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Table 1. Mean percent survival of *H. Azteca* and concentrations of organophosphate pesticides, and toxic units from Phase 1 TIE treatments conducted on 12/4/03. NA indicates not analyzed

Treatment	Percent Sample 0%	10%	25%	50%	100%	Chlorpyrifos (µg/L)	Diazinon (µg/L)
Initial Test	100	100	80	44	8		
48-Hour Results							
Baseline 15C	100			0	0	NA	NA
Baseline 23C	93			0	0	NA	NA
C18 Column	100			67	0	NA	NA
C18 Eluate	100			87	40	NA	NA
PBO	100			13	0	NA	NA
Enzyme	100			47	0	NA	NA
Enzyme/PBO	100			13	0	NA	NA
96-Hour Results							
Baseline 15C	100			0	0	NA	NA
Baseline 23C	87			0	0	NA	NA
C18 Column	93			0	0	NA	NA
C18 Eluate	93			60	0	NA	NA
PBO	93			0	0	NA	NA
Enzyme	100			0	0	NA	NA
Enzyme/PBO	93			0	0	NA	NA

Table 2. Water quality measurements for Phase 1 TIE treatments conducted on 12/4/03. ND indicates non-detect.

Treatment	pH	Dissolved Oxygen (mg/L)	Conductivity (µS/cm)	Total Ammonia (mg/L)	Un-ionized Ammonia (mg/L)
Baseline	7.92	6.09	1884	2.2	0.087
C18 Column	8.10	8.34	1859	2.6	0.152
C18 Eluate	8.26	8.10	677	ND	ND
PBO	7.88	6.21	1842	2.4	0.087
Enzyme	7.89	6.36	1788	16.8	0.619
Enzyme/PBO	7.89	6.40	1788	15.7	0.579

Appendix B-3

Ingram Creek at River Road
September 13, 2004 sampling event

Toxicity Identification Evaluation Results

Region 5 – Combined Stations 03-ICARR-014 and 03-ICARR-015

Surface Water Ambient Monitoring Program

By the University of California, Davis - Department of Environmental Toxicology

Marine Pollution Studies Laboratory
34500 Coast Route One, Granite Canyon
Monterey, CA 93940

Submitted to the Central Valley Regional Water Quality Control Board – Sacramento Region 5

February 2005

Introduction

Two sediment samples were submitted as part of the West Side Coalition Agricultural waiver monitoring program and were collected by of John Hansen from Del Puerto Water District and Chris Linneman from Summers engineering. The West Side Coalition collected additional sample for the Surface Water Ambient Monitoring Program (SWAMP) for combined toxicity identification (TIE) analysis. Prior to submittal, solid-phase toxicity tests were performed by Pacific Eco Risk (Martinez, CA), and significant toxicity to *Hyalella azteca* was observed. Upon receipt, the sediments were combined and homogenized. After interstitial water was extracted, a TIE with *Hyalella azteca* was conducted using established testing protocols. TIEs are designed to proceed in three phases. The purpose of a Phase 1 TIE is to characterize the cause of toxicity. Information from the Phase 1 characterization may then be used in subsequent Phase 2 (identification) and Phase 3 (confirmation) TIEs. Based on the results of initial toxicity tests, an abbreviated Phase 1 TIE was conducted to investigate the causes of toxicity. The TIE did not utilize the normal suite of treatments because of minimal sample availability. This report presents the data obtained from the TIE, including the mean percent survival of amphipods after exposure to various TIE treatments, water quality measurements of test solutions, and copies of the original data sheets and quality assurance forms.

Methods

Sample Handling

The sediment samples were collected on September 13, 2004, the field sampling was performed by Chris Linneman from Summers Engineering and John Hansen from Del Puerto Water District. After initial solid phase testing at Pacific Ecorisk, the samples were transported on ice and in the dark to the Marine

Pollution Studies Laboratory at Granite Canyon for initial interstitial water toxicity testing. Interstitial water was extracted from the sample on October 27, and an initial test started on October 28, 2004. After the termination of the initial test, the TIE was initiated on November 5, 2004.

TIE Methods

The following Phase 1 TIE treatments were performed on a dilution series of each sample (US EPA 1991). Sample concentrations in the initial test were 0 (treatment blank), 25, and 100%. The treatment blank was control water that underwent the same manipulation as the sample.

Treatments:

• Baseline - Toxicity test on un-manipulated sample. Concentrations were chosen to bracket the effect concentration of the sample and might differ from initial test.

• C8 Column - The C8 Column is designed to remove non-polar organic compounds from the sample. In the manipulation, reverse phase liquid chromatography is applied to extract nonionic organic toxicants from the aqueous sample. Column can be eluted with methanol and resulting eluate tested to determine if substances removed by the column are indeed toxic.

• EDTA (Disodium Ethylenediaminetetraacetic acid) - EDTA is an organic chelating agent that preferentially binds with divalent metals, such as copper, nickel, lead, zinc, cadmium, mercury, and other transition metals to form non-toxic complexes. It will not complex with anionic forms of metals such as selenids, chromates and hydrochromates.

• PBO (Piperonyl Butoxide) - PBO is a metabolic inhibitor that removes the toxicity associated with metabolically activated pesticides such as diazinon and chlorpyrifos. An increase of toxicity with the PBO treatments can indicate the presence of non-metabolically activated compounds such as pyrethroid pesticides.

• Carboxylesterase Enzyme - Porcine carboxylesterase was added to the sample to break down suspected pyrethroid pesticides (Wheelock et al. 2004).

Exposures were conducted in 20 mL glass scintillation vials (3 replicates) containing 15 mL treated sample and five amphipods. Acute exposures were conducted for 96 hours, following US EPA 1993.

Physical and Chemical Measurements

Water quality parameters of dissolved oxygen, pH and conductivity were measured using a Hach SensION© selective ion meter with appropriate electrodes; and ammonia was measured using a Hach 2010 spectrophotometer. Temperature was measured using a continuously recording thermograph and thermometer. Concentrations of the organophosphate pesticides chlorpyrifos and diazinon were measured using enzyme-linked immunosorbent assays (ELISA, Strategic Diagnostics Inc, Newark, DE). Water quality parameters were only measured on 100% Baseline sample because of lack of sample.

Data Interpretation

Treatment blanks were evaluated to determine if sample manipulations added toxic artifacts. Treatment data were then compared to one another based on organism response.

Results and Discussion

The initial test was significantly toxic to *H. azteca* at the 10% concentration in a 96-hour acute exposure (LC50 = 5.8%, Table 1). There was no toxicity observed in the treatment blanks. Water quality parameters were all within the tolerance limits of the test organism. Concentrations of chlorpyrifos and diazinon were below detection limits (Table 1).

At 96 hours the Baseline treatment had complete mortality in both the 25% and 100% concentrations. The only treatment that reduced toxicity was the Enzyme, indicating the cause of toxicity was a pyrethroid pesticide, or a combination of pyrethroids (Table 1). Additional evidence for pyrethroid toxicity is in the results of the PBO treatment. At 48 hours the toxicity of the PBO treatment was higher than Baseline indicating the signal was being increased by the addition of PBO. The C8 Column did not reduce toxicity, but it did bind some non-polar organic contaminants because the Column Eluate returned significant toxicity to clean dilution water. The sediment and the methanol extract from the C8 Column were analyzed for pyrethroid pesticides by the Water Pollution Control Laboratory (Rancho Cordova, CA).

The sediment contained cyfluthrin, esfenvalerate/fenvalerate, and lambda cyhalothrin (Table 2). The concentration of lambda cyhalothrin was more than four times the mean sediment LC50 reported by Amweg et al. (In Press). Although the methanol extract from the column eluate treatment returned toxicity to clean water, chemical analysis of the extract did not find any pyrethroid pesticides (Table 2).

Because of the minimal sample provided, additional sediment chemistry could not be conducted, yet additional analysis of the methanol extract is still feasible providing the chemistry laboratory has leftover extract. The high concentration of lambda cyhalothrin strongly suggests the cause of toxicity to be a pyrethroid, but additional factors could be contributing to toxicity.

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Wheelock CE, Miller JL, Miller MJ, Gee SJ, Shan G, Hammock BD. 2004. Development of toxicity identification evaluation procedures for pyrethroid detection using esterase activity. *Environ Toxicol Chem*. 23: 2699-2708.

Table 1. Mean percent survival of *H. azteca* and concentrations of organophosphate pesticides from Phase 1 TIE treatments conducted on 11/5/04. NA indicates not analyzed. ND indicates not detected.

	Percent Sample					Chlorpyrifos	Diazinon
Treatment	0%	10%	25%	50%	100%	(μg/L)	(μg/L)
Initial Test	100	13	7	0	0	ND	ND
48-Hour Results							
Baseline	100		73		0	NA	NA
C8 Column	100		33		0	NA	NA
C8 Eluate	100		60		73	NA	NA
EDTA	100		53		7	NA	NA
PBO	100		27		0	NA	NA
Enzyme	100		100		93	NA	NA
96-Hour Results							
Baseline	100		0		0	NA	NA
C8 Column	100		0		0	NA	NA
C8 Eluate	100		27		0	NA	NA
EDTA	100		0		0	NA	NA
PBO	100		0		0	NA	NA
Enzyme	100		100		73	NA	NA

Table 2. Pyrethroid concentrations in sediment and methanol extract, and mean sediment LC50 values from Amweg et al. (In Press). ND indicates not detected. NR indicates not reported.

Pyrethroid	Sediment ng/g dry wt.	Methanol Extract	Sediment LC50 ng/g dry wt.
Bifenthrin	ND	ND	4.4
Cyfluthrin	3.21	ND	14.2
Cypermethrin	ND	ND	NR
Esfenvalerate/Fenvalerate	5.47	ND	42.2
Lambda (Cyhalothrin)	25.2	ND	5.8
Permethrin	ND	ND	206.3

Appendix B-4

Hospital Creek at River Road
March 30, 2005 sampling event

Toxicity Identification Evaluation Results
Region 5 – Hospital Creek at River Road – 541STC042
Surface Water Ambient Monitoring Program

By the University of California, Davis - Department of Environmental Toxicology

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Submitted to the Central Valley Regional Water Quality Control Board – Sacramento Region 5

September 2005

Introduction

A sediment sample from Hospital Creek at River Road was submitted as part of the West Side Coalition Agricultural waiver-monitoring program and was collected by Chris Linneman from Summers engineering. The West Side Coalition collected additional sample for the Surface Water Ambient Monitoring Program (SWAMP) for toxicity identification (TIE) analysis. Pacific Eco Risk (Martinez, CA) performed an initial solid-phase toxicity test prior to submittal, and significant toxicity to *Hyalella azteca* was observed. Upon receipt, interstitial water was extracted and an initial screening test was initiated.

TIEs are designed to proceed in three phases. The purpose of a Phase 1 TIE is to characterize the cause of toxicity. Information from the Phase 1 characterization may then be used in subsequent Phase 2 (identification) and Phase 3 (confirmation) TIEs. The results of initial toxicity tests demonstrated that interstitial water was not significantly toxic to *H. azteca*; so solid-phase TIE protocols were used. This report presents the data obtained from the TIE, including the mean percent survival of amphipods after exposure to various TIE treatments, water quality measurements of test solutions, and copies of the original data sheets and quality assurance forms.

Methods

Sample Handling

The sediment samples were collected on March 30, 2005. After initial solid phase testing at Pacific Eco Risk, the samples were transported on ice and in the dark to the Marine Pollution Studies Laboratory at Granite Canyon for initial interstitial water toxicity testing. Interstitial water was extracted from the sample on April 7, and an initial test started on April 8, 2005. After the termination of the initial test, the solid-phase TIE was initiated on April 29, 2005.

TIE Methods

The following solid-phase TIE treatments were performed on undiluted sediment. Treatment blanks consisted of laboratory formulated sediment that underwent the same treatment as the sample. Formulated sediment was prepared using equal parts Salinas River, California reference site sediment and clean, kiln-dried sand (#60, RMC Pacific Materials, Monterey, CA, USA). The sediment was amended with 1.5% organic peat moss (Uni-Gro, Chino, CA, USA). One kilogram (dry weight) of formulated sediment was prepared by combining 500g reference sediment, 500g sand, and 7.5g peat with 350 mL clean dilution water. Phase I TIE treatments consisted of Ambersorb addition to the sediment, carboxylesterase addition to the overlying water, and piperonyl butoxide addition to the overlying water. The baseline and enzyme treatments were also performed at a colder temperature to determine if pyrethroids or DDT caused toxicity. Phase II TIE procedures consisted of separating the Ambersorb from the sediment, extracting it with solvent, and spiking control water with the methanol eluate as a toxicity add-back procedure.

Treatment

• Baseline - Toxicity test on un-manipulated sample. Five 250-mL replicate beakers each containing approximately 50g sediment and 200 mL clean dilution water.

• Ambersorb 563® (Rohm and Haas, Spring House, PA, USA), a carbonaceous, non-polar resin, was prepared by rinsing it thoroughly with Nanopure® water. Ten percent Ambersorb by wet weight was added to sediment (Kosian et al. 1999, West et al. 2001). Treated sediment was homogenized for 24 hours on the roller apparatus, and then loaded into exposure chambers. A dilution blank was created by combining test sediment with 5% formulated sediment, and an Ambersorb blank was created by adding 5% Ambersorb to formulated sediment. At test termination the sediment was sieved through a series of screens ranging from 250-400 μm to retain the Ambersorb. The Ambersorb was then eluted by loading a column with approximately 7.5g resin and pumping 10 mL of acetone through the column at a rate of 1 mL per minute. Post-column acetone was collected in a 50 mL beaker and evaporated to a final volume of one mL. The final volume was combined with 100 mL clean dilution water to create the eluate sample for toxicity testing with *H. azteca*. An Ambersorb elution blank was prepared by performing the above treatments on Ambersorb that had been combined with formulated sediment. A 1% acetone blank was also tested.

• Piperonyl butoxide (Sigma-Aldrich, St. Louis, MO) is used to block the metabolic activation of acetylcholinesterase-inhibiting organophosphate pesticides (Ankley et al. 1991). It is also a potent synergist of pesticide toxicity, because it inhibits their metabolism (Ware 1989, Kakko et al. 2000). The PBO treatment contained 500 μg/L of PBO in the water overlying the sediment. Decreased toxicity with the addition of PBO suggests the presence of organophosphate pesticides. Increased toxicity with the addition of PBO suggests the presence of pyrethroids.

• The enzyme carboxylesterase (Sigma-Aldrich, St. Louis, MO) hydrolyzes ester-containing compounds such as pyrethroids to their corresponding acid and alcohol, which are generally not toxic (Wheelock et al. 2004). Carboxylesterase (500x) was added to the overlying water on the day of test initiation, six hours prior to the addition of amphipods. This allowed for interaction between the enzyme and pyrethroids. The enzyme was added based on units of activity. One 'x' of enzyme activity equals 0.0025 units of enzyme per mL of sample, therefore at 500x, 1.25 units per mL were added. Enzyme strength is unique for each lot purchased (Wheelock et al. 2004). A combination treatment of enzyme and PBO was also conducted to help resolve toxicity due to combinations of organophosphate and pyrethroid pesticides. Enzyme was added and allowed to interact with the overlying water for six hours before the addition of PBO, and then amphipods.

• Additional baseline and enzyme treatments were conducted at 15ºC because the toxicity of some organochlorine and pyrethroid pesticides increases with decreasing temperature (Ware 1989).

Physical and Chemical Measurements

Water quality parameters of dissolved oxygen, pH and conductivity were measured using a Hach SensION© selective ion meter with appropriate electrodes; and ammonia was measured using a Hach 2010 spectrophotometer. Temperature was measured using a continuously recording thermograph and thermometer. Concentrations of the organophosphate pesticides chlorpyrifos and diazinon were measured using enzyme-linked immunosorbent assays (ELISA, Strategic Diagnostics Inc, Newark, DE). Water quality parameters were only measured on 100% Baseline sample because of lack of sample.

Data Interpretation

Treatment blanks were evaluated to determine if sample manipulations added toxic artifacts. Treatment data were then compared to one another based on organism response.

Results and Discussion

Survival in the initial interstitial water test was 73%. This toxicity signal was not considered strong enough to pursue an interstitial water TIE. Interstitial water was also analyzed for chlorpyrifos and diazinon using ELISA. Concentrations of both organophosphates were below reporting limits.

Survival in the baseline sediment exposure was 22 percent, and all treatment blanks had acceptable survival (Table 1). Survival was increased with the addition of Ambersorb to the sediment and with the addition of enzyme to the overlying water. There was some dilution effect created by adding 10%

amendment to the sediment, but not enough to account for the reduction in toxicity by the addition of Ambersorb. Treatments that contained PBO exhibited complete mortality. Toxicity was also increased when the exposure was conducted at 15ºC (6% survival vs. 22% survival in the 23ºC baseline). The 15ºC enzyme treatment reduced toxicity, but to a lesser extent than the 23ºC enzyme treatment.

Increased survival with Ambersorb indicates the cause of toxicity was organic, and the increase of survival with enzyme indicates the cause of toxicity was a pyrethroid. Because the enzyme is a protein, it can also reduce the bioavailability of contaminants through adsorption. We did not use a protein control in this experiment to account for the effects of protein binding. Increased toxicity at 15ºC can indicate toxicity due to pyrethroids or DDT.

Ambersorb was isolated from the test sediment at the termination of the exposure and eluted with acetone. When the acetone was added back to clean dilution water it caused significant toxicity indicating an organic was removed from the sediment by the Ambersorb and was successfully added back to clean dilution water (Table 1).

Based on the toxicity results, the sediment and acetone extract were analyzed for organochlorine pesticides and pyrethroids. The sediment contained es-fenvalerate, lambda-cyhalothrin, DDT, and endosulfan (Table 2). The acetone extract contained DDT only. The concentrations of the chemicals in the sediment were not higher than known effects thresholds, but the concentrations of DDT metabolites in the acetone extract exceeded the LC50 values of Hoke et al. (1994) after dilution.

Implicating the class of chemicals responsible for the cause of sediment toxicity requires multiple lines of evidence that include the results of TIE treatments, as well as chemical analyses. The reduction of toxicity with the addition of enzyme and the increase in toxicity with the addition of PBO suggests the cause of toxicity was pyrethroids, but the sediment concentrations of six pyrethroids were well below the published LC50 values. The enzyme might have reduced toxicity by binding chemicals in its protein matrix, but we are not sure because we did not include a protein control. Also, there is some evidence that PBO increases toxicity caused by DDT (Brandt et al. 2002), but we are unaware of specific studies exposing *H. azteca* to mixtures of PBO and DDT. Although the concentration of total DDT in the sediment was below the published LC50 value, the concentrations of DDT metabolites in the acetone eluate were above water-only LC50 values after dilution. The increase of toxicity at low temperature is consistent with DDT toxicity as well as that of pyrethroids, but the fact that only DDT was found in the acetone eluate provides compelling evidence that DDT is the main cause of toxicity with pyrethroids perhaps playing a lesser role.

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Table 1. Mean percent survival and standard deviation (SD) of *H. azteca* in solid-phase TIE treatments and acetone eluate treatments.

	Solid-Phase Treatments		Acetone Eluate Treatments	
Treatment (23ºC)	Mean	SD	Mean	SD
Baseline Sediment	22	29		
Sediment (10% Ambersorb)	90	10	0	0
Control (10% Ambersorb)	98	4	100	0
Sediment (Enzyme)	96	5		
Control (Enzyme)	94	9		
Sediment (PBO)	0	0		
Sediment (Enzyme/PBO)	0	0		
Control (Enzyme/PBO)	100	0		
Sediment (10% Control)	48	8		
Control	98	4	93	12
Treatment (15ºC)	Mean	SD		
Baseline Sediment	6	9		
Sediment (Enzyme)	46	27		
Control	96	9		

Table 2. Chemical concentrations in sediment and diluted acetone extract, and respective sediment and water LC50 values. ND indicates not detected. NR indicates not reported.

Chemical	Sediment ng/g dry wt.	Sediment LC50 ng/g dry wt.	Acetone Extract µg/L	Water LC50 µg/L
DDD (p,p’)	4.12	-	0.64	0.19 [a]
DDE (p,p’)	35.7	-	3.22	1.66 [a]
DDT (o,p’)	4.22	-	1.93	0.07 [a]
DDT (p,p’)	13.7	-	0.80	-
Total DDT	57.74	371 µg/g oc [b]	NR	-
Es-fenvalerate-2	0.32	42.2 [c]	ND	-
Lambda-cyhalothrin-2	0.18	5.8 [c]	ND	-

a Hoke et al. 1994, b Nebeker et al. 1989, c Amweg et al. 2004.

Table 3. Water quality measurements for Hospital Creek solid-phase TIE. ND indicates non detect. NA indicates not analyzed.

	pH		Dissolved Oxygen (mg/L)		Conductivity (µS/cm)		Total Ammonia (mg/L)		Un-ionized Ammonia (mg/L)	
Treatment (23ºC)	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Baseline Sediment	8.32	8.70	8.18	8.21	687	1062	0.2	ND	0.019	ND
Sediment (10% Ambersorb)	8.34	8.66	8.05	8.19	701	914	0	ND	0.000	ND
Control (10% Ambersorb)	8.11	8.61	6.65	7.69	692	1344	1.6	12.4	0.096	2.074
Sediment (Enzyme)	8.24	8.10	6.93	7.20	645	716	0.8	ND	0.063	ND
Control (Enzyme)	8.18	8.10	7.08	7.18	632	744	0.7	ND	0.049	ND
Sediment (PBO)	8.32	8.62	7.81	8.21	645	8.40	0	ND	0.000	ND
Sediment (Enzyme/PBO)	8.34	8.12	7.18	7.37	617	691	0.6	ND	0.058	ND
Control (Enzyme/PBO)	8.05	8.43	5.17	7.09	901	1128	2.0	18.5	0.105	2.168
Sediment (10% Control)	8.25	8.62	7.61	8.27	696	778	ND	ND	ND	ND
Control	7.96	8.58	5.67	7.67	1035	1393	2.3	13.5	0.099	2.132
Treatment (15ºC)										
Baseline Sediment	8.12	8.28	9.37	8.15	631	862	ND	0.2	ND	0.017
Sediment (Enzyme)	8.17	8.00	8.95	5.32	626	740	1.3	14.0	0.045	0.329
Control	8.07	8.36	9.17	8.46	811	1343	0.5	7.6	0.027	0.771

Appendix B-5

Grayson Road Drain
June 15, 2005 sampling event

Toxicity Identification Evaluation Results

Region 5 – Grayson Drain – 541STC030

Surface Water Ambient Monitoring Program

By the University of California, Davis - Department of Environmental Toxicology

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Submitted to the Central Valley Regional Water Quality Control Board – Sacramento Region 5

February 2006

Introduction

A sediment sample from Grayson Drain was submitted as part of the Surface Water Ambient Monitoring Program (SWAMP) and was collected by Jay Rowan of the Central Valley Regional Water Quality Control Board. A toxicity identification evaluation (TIE) was initiated on July 8, 2005 with a screening of interstitial water toxicity.

TIEs are designed to proceed in three phases. The purpose of a Phase I TIE is to characterize the cause of toxicity. Information from the Phase I characterization may then be used in subsequent Phase II (identification) and Phase III (confirmation) TIEs. The results of initial toxicity tests demonstrated that interstitial water was not significantly toxic to *H. azteca*; so solid-phase TIE protocols were used. This report presents the data obtained from the TIE, including the mean percent survival of amphipods after exposure to various TIE treatments, water quality measurements of test solutions, and copies of the original data sheets and quality assurance forms.

Methods

Sample Handling

The sediment was collected on June 15, 2005 and the initial solid-phase test was conducted on June 21, 2005. Survival in the initial test was 38%. Interstitial water was extracted from the sample on July 7, and an initial test started on July 8, 2005. After the termination of the initial test, the solid-phase TIE was initiated on August 5, 2005.

TIE Methods

The following solid-phase TIE treatments were performed on undiluted sediment. Treatment blanks consisted of laboratory formulated sediment that underwent the same treatment as the sample. Formulated sediment was prepared using equal parts Salinas River, California reference site sediment and clean, kiln-dried sand (#60, RMC Pacific Materials, Monterey, CA, USA). The sediment was amended with 0.75% organic peat moss (Uni-Gro, Chino, CA, USA). One kilogram (dry weight) of formulated sediment was prepared by combining 500g reference sediment, 500g sand, and 7.5g peat with 350 mL clean dilution water. Phase I TIE treatments consisted of Ambersorb addition to the sediment and carboxylesterase addition to the overlying water. The baseline and enzyme treatments were also performed at a colder temperature to determine if pyrethroids or DDT caused toxicity. Phase II TIE procedures consisted of separating the Ambersorb from the sediment, extracting it with solvent, and spiking control water with the acetone eluate as a toxicity return procedure.

Treatment

• Baseline - Toxicity test on un-manipulated sample. Five 250-mL replicate beakers each containing approximately 50g sediment and 200 mL clean dilution water.

• Ambersorb 563® (Rohm and Haas, Spring House, PA, USA), a carbonaceous, non-polar resin, was prepared by rinsing it thoroughly with Nanopure® water. Ten percent Ambersorb by wet weight was added to sediment (Kosian et al. 1999, West et al. 2001). Treated sediment was homogenized for 24 hours on a roller apparatus, then loaded into exposure chambers. A dilution blank was created by combining test sediment with 10% formulated sediment, and an Ambersorb blank was created by adding 10% Ambersorb to formulated sediment. At test termination the sediment was sieved through a series of screens ranging from 250-400 μm to retain the Ambersorb. The Ambersorb was then eluted by loading a column with approximately 7.5g resin and pumping 10 mL of acetone through the column at a rate of 1 mL per minute. Post-column acetone was collected in a 50 mL beaker and evaporated to a final volume of one mL. The final volume was combined with 100 mL clean dilution water to create the eluate sample for toxicity testing with *H. azteca*. An Ambersorb elution blank was prepared by performing the above treatments on Ambersorb that had been combined with formulated sediment. A 1% acetone blank was also tested.

• The enzyme carboxylesterase (Sigma-Aldrich, St. Louis, MO) hydrolyzes ester-containing compounds such as pyrethroids to their corresponding acid and alcohol, which are generally not toxic (Wheelock et al. 2004). Carboxylesterase (500x) was added to the overlying water on the day of test initiation, six hours prior to the addition of amphipods. This allowed for interaction between the enzyme and pyrethroids. The enzyme was added based on units of activity. One ‘x’ of enzyme activity equals 0.0025 units of enzyme per mL of sample, therefore at 500x, 1.25 units per mL were added. Enzyme strength is unique for each lot purchased (Wheelock et al. 2004). To control for the binding of contaminants to the protein base of the enzyme, a separate set of replicates was treated with bovine serum albumin (BSA). Reduction of toxicity by the enzyme, and not the BSA, would confirm the presence of pyrethroids. The enzyme and protein treatments were given daily booster shots of BSA and carboxylesterase.

• Additional baseline and enzyme treatments were conducted at 15ºC because the toxicity of some organochlorine and pyrethroid pesticides increases with decreasing temperature (Ware 1989).

Physical and Chemical Measurements

Water quality parameters of dissolved oxygen, pH and conductivity were measured using a Hach SensION© selective ion meter with appropriate electrodes; and ammonia was measured using a Hach 2010 spectrophotometer. Temperature was measured using a continuously recording thermograph and thermometer. Concentrations of the organophosphate pesticides chlorpyrifos and diazinon were measured using enzyme-linked immunosorbent assays (ELISA, Strategic Diagnostics Inc, Newark, DE).

Data Interpretation

Treatment blanks were evaluated to determine if sample manipulations added toxic artifacts. Treatment data were then compared to one another based on organism response.

Results and Discussion

Survival in the initial interstitial water test was 87%. This toxicity signal was not strong enough to pursue an interstitial water TIE. Interstitial water was also analyzed for chlorpyrifos and diazinon using ELISA. The concentration of diazinon was below reporting limits, but the concentration of chlorpyrifos was 0.122 µg/L, which was slightly higher than the LC50 for *H. azteca* (Phipps et al. 1995). Because there was no apparent interstitial water toxicity, it is assumed that the chlorpyrifos was not bioavailable in the interstitial water exposure.

Two solid-phase TIEs were conducted. The first TIE included the Ambersorb treatment and the 15ºC treatments. The second TIE included the carboxylesterase and BSA treatments. Survival in the first baseline sediment exposure was 10%, and all treatment blanks had acceptable survival (Table 1). Survival was increased by the addition of Ambersorb to the sediment. The cold temperature baseline survival was 2%, and the addition of enzyme increased survival to 22%. Baseline survival in the second TIE was 26%. Addition of the enzyme and the BSA both significantly increased survival (Table 2).

Based on the solid-phase Phase I TIE results, the cause of toxicity was characterized as an organic, but the results of the enzyme and BSA treatments rule out pyrethroid toxicity. Increased toxicity at cold temperature indicates that DDT and its metabolites might be contributing to toxicity.

Ambersorb was isolated from the test sediment at the termination of the exposure and eluted with acetone. When the acetone was added back to clean dilution water it caused significant toxicity indicating an organic was removed from the sediment by the Ambersorb and was successfully returned to clean dilution water (Table 1). Based on the toxicity results, the sediment and acetone extract were analyzed for organochlorine pesticides and pyrethroids. The sediment contained chlorpyrifos and several organochlorine pesticides, but no pyrethroids (Table 3). The acetone extract also contained chlorpyrifos and organochlorine pesticides. The extract results are presented as the concentration of chemical after reconstitution. The concentrations of the chemicals in the sediment were not higher than known effects thresholds, but the concentrations of chlorpyrifos and DDT metabolites in the acetone extract exceeded published LC50 values (Table 3). When compared to the consensus-based probable effects concentrations (PEC) of MacDonald et al. (2000), the concentration of sum DDE was greater than the PEC. All water quality parameters were within acceptable limits for *H. azteca* (Table 4).

Implicating the class of chemicals responsible for the cause of sediment toxicity requires multiple lines of evidence that include the results of TIE treatments, as well as chemical analyses. Reduction of toxicity with Ambersorb, and the subsequent return of toxicity with the Ambersorb eluate characterize the cause of toxicity as organic. Increase of toxicity at cold temperature suggests the cause is either a pyrethroid or DDT, but the removal of toxicity by the BSA as well as the enzyme ruled out toxicity caused by pyrethroids.

The sediment contained chlorpyrifos, but at a concentration much lower than the published LC50. Similarly, the concentration of total DDT, corrected for organic carbon, was also much lower than the published LC50, but the concentration of sum DDE was above the consensus-based PEC of 31.3 ng/g (MacDonald et al. 2000). Conversely, concentrations of chlorpyrifos and DDT metabolites in the Ambersorb eluate were greater than published LC50s. It should be noted that the volume of solvent extract and the volume of water used to reconstitute the solvent was arbitrary, and was used simply to indicate the removal of the contaminant by the Ambersorb. Although these volumes were chosen for convenience, the toxicity results and chemical analyses of the eluate provide additional lines of evidence for the cause of toxicity. The presence of chlorpyrifos and DDT metabolites at toxic concentrations in the Ambersorb eluate suggest that these chemicals are the primary cause of toxicity in this sediment sample.

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Wheelock CE, Miller JL, Miller MJ, Gee SJ, Shan G, Hammock BD. 2004. Development of toxicity identification evaluation procedures for pyrethroid detection using esterase activity. *Environ Toxicol Chem*. 23: 2699-2708.

Table 1. Results of first solid-phase TIE. Mean percent survival and standard deviation (SD) of *H. azteca* in solid-phase TIE treatments and acetone eluate treatments.

	Solid-Phase Treatments		Acetone Eluate Treatments	
Treatment (23ºC)	Mean	SD	Mean	SD
Baseline Sediment	10	7		
Sediment (10% Ambersorb)	52	23	0	0
Control (10% Ambersorb)	86	9	87	23
Sediment (10% Control)	8	18		
Control	100	0	100	0
Treatment (15ºC)	Mean	SD		
Baseline Sediment	2	4		
Sediment (Enzyme)	22	27		
Control (Enzyme)	74	18		
Control	92	11		

Table 1. Results of second solid-phase TIE. Mean percent survival and standard deviation (SD) of *H. azteca* in solid-phase TIE treatments.

	Solid-Phase Treatments	
Treatment	Mean	SD
Baseline Sediment	26	17
Sediment (Enzyme)	96	5
Control (Enzyme)	100	0
Sediment (BSA)	100	0
Control (BSA)	100	0
Control	100	0

Table 3. Chemical concentrations in sediment and diluted acetone extract, and respective sediment and water LC50 values. ND indicates not detected. NR indicates not reported.

Chemical	Sediment ng/g dry wt.	Sediment LC50 ng/g dry wt.	Acetone Extract μg/L	Water LC50 μg/L
Chlorpyrifos	10.7	399 [a]	0.792	0.086 [b]
Chlordane, cis-	3.25		0.312	
Chlordane, trans-	<RL		0.342	
Nonachlor, cis-	ND		0.179	
Nonachlor, trans-	2.08		0.276	
DDD (o,p')	ND		0.634	
DDD (p,p')	26.5		1.60	0.19 [c]
DDE (o,p')	3.21		0.151	
DDE (p,p')	134		2.04	1.66 [c]
DDT (o,p')	17.7		1.34	0.07 [c]
DDT (p,p')	ND		1.60	
Total DDT	24.9 μg/g oc	371 μg/g oc [d]	NR	
Dieldrin	ND		0.390	
Endrin	<RL		0.459	
Total Organic Carbon	0.73%			

[a] Brown et al. 1997, [b] Phipps et al. 1995, [c] Hoke et al. 1994, [d] Nebeker et al. 1989.

Table 4. Water quality measurements for solid-phase TIEs. ND indicates non-detect. NA indicates not analyzed.

TIE 1	pH		Dissolved Oxygen (mg/L)		Conductivity (µS/cm)		Total Ammonia (mg/L)		Un-ionized Ammonia (mg/L)	
Treatment (23ºC)	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Baseline Sediment	8.3	8.74	8.77	8.47	689	925	0.2	ND	0.018	ND
Sediment (10% Ambersorb)	8.35	8.98	8.63	8.58	723	978	0.1	ND	0.010	ND
Control (10% Ambersorb)	8.32	8.95	8.54	8.56	1076	1517	1.7	2.3	0.159	0.702
Sediment (10% Control)	8.37	8.97	8.58	8.58	773	972	0.5	ND	0.052	ND
Control	8.28	8.99	8.15	8.23	1119	1359	2.2	8.2	0.189	2.667
Treatment (15ºC)										
Baseline Sediment	8.18	8.89	8.92	9.19	688	790	0.1	ND	0.004	ND
Sediment (Enzyme)	8.22	8.67	9.09	8.83	671	785	0.6	2.3	0.026	0.260
Control (Enzyme)	8.19	8.82	8.98	8.81	991	1456	2	17.9	0.081	2.734
Control	8.15	9	9.09	9.28	1083	1609	1.9	7.1	0.071	1.522
TIE 2	pH		Dissolved Oxygen (mg/L)		Conductivity (µS/cm)		Total Ammonia (mg/L)		Un-ionized Ammonia (mg/L)	
Treatment (23ºC)	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Baseline Sediment	8.49	8.43	8.46	8.21	704	915	0.5	0	0.066	0.000
Sediment (Enzyme)	8.48	8.35	8.5	7.7	883	929	0.4	16	0.052	1.591
Control (Enzyme)	8.29	8.47	7.29	7.28	1070	1571	2.6	32.6	0.228	4.142
Sediment (BSA)	8.47	8.28	8.4	5.57	677	983	0.5	17.8	0.064	1.529
Control (BSA)	8.24	8.45	7.52	6.64	1012	1513	1.3	34.2	0.103	4.174
Control	8.25	8.6	7.29	8.1	1128	1498	2.2	9.4	0.177	1.543

Appendix B-6

Westly Wasteway near Cox Road and Del Puerto Creek near Cox Road
October 10, 2005 sampling event

Toxicity Identification Evaluation Results
Region 5 – DPCCR (541STC533) and WWNCR (541STC029)
Surface Water Ambient Monitoring Program

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Introduction

Two sediment samples were submitted as part of the West Side Coalition Agricultural waiver monitoring program and were collected by John Hansen from Del Puerto Water District. The West Side Coalition collected additional sample for the Surface Water Ambient Monitoring Program (SWAMP) for toxicity identification (TIE) analysis. Prior to submittal, solid-phase toxicity tests were performed by Pacific Eco Risk (Martinez, CA), and significant toxicity to *Hyalella azteca* was observed.

Sediment from stations DPCCR (541STC533) and WWNCR (541STC029) were received at the Marine Pollution Studies Laboratory (MPSL) on November 14, 2005. On November 18, 2005 initial toxicity tests on sediment and interstitial water were initiated using established testing protocols.

TIEs are designed to proceed in three phases. The purpose of a Phase I TIE is to characterize the cause of toxicity. Information from the Phase I characterization may then be used in subsequent Phase II (identification) and Phase III (confirmation) TIEs. The results of initial toxicity tests demonstrated that both interstitial water and sediment were significantly toxic to *H. azteca*. Phase I and II TIEs were conducted on both matrices. This report presents the data obtained from the TIEs, including the mean percent survival of amphipods after exposure to various TIE treatments, water quality measurements of test solutions, and copies of the original data sheets and quality assurance forms.

Methods

Sample Handling

The sediment was collected on October 10, 2005. After receiving the samples from Pacific Ecorisk, the initial solid-phase tests were conducted on November 18, 2005. Once the magnitude of the initial toxicity was determined, interstitial water TIEs were initiated on December 16, 2005 and solid-phase TIEs were initiated on December 17, 2005.

TIE Methods

The following solid-phase TIE treatments were performed on undiluted sediment. Treatment blanks consisted of laboratory formulated sediment that underwent the same treatment as the sample. Formulated sediment was prepared using equal parts Salinas River, California reference site sediment and clean, kiln-dried sand (#60, RMC Pacific Materials, Monterey, CA, USA). The sediment was amended with 0.75% organic peat moss (Uni-Gro, Chino, CA, USA). One kilogram (dry weight) of formulated sediment was prepared by combining 500g reference sediment, 500g sand, and 7.5g peat with 350 mL clean dilution water. Phase I TIE treatments consisted of additions of amendments to the sediment, or treatments of the overlying water. Sediment amendments included Ambersorb and SIR-300. Overlying water treatments consisted of addition of carboxylesterase enzyme, bovine serum albumin (BSA), and piperonyl butoxide (PBO). The baseline treatment was also performed at a colder temperature to determine if pyrethroids or DDT caused toxicity. Phase II TIE procedures consisted of separating the Ambersorb from the sediment, extracting it with solvent, and spiking control water with the acetone eluate as a toxicity return procedure.

Solid-Phase Treatments

• Baseline - Toxicity test on un-manipulated sample. Five 250-mL replicate beakers each containing approximately 50g sediment and 200 mL clean dilution water.

• Ambersorb 563® (Rohm and Haas, Spring House, PA, USA), a carbonaceous, non-polar resin, was prepared by rinsing it thoroughly with Nanopure® water. Ten percent Ambersorb by wet weight was added to sediment (Kosian et al. 1999, West et al. 2001). Treated sediment was homogenized for 24 hours on a roller apparatus and loaded into exposure chambers. A dilution blank was created by combining test sediment with 10% formulated sediment, and an Ambersorb blank was created by adding 10% Ambersorb to formulated sediment. At test termination the sediment was sieved through a series of screens ranging from 250-400 μm to retain the Ambersorb. The Ambersorb was then eluted by loading a column with approximately 7.5g resin and pumping 10 mL of acetone through the column at a rate of 1 mL per minute. Post-column acetone was collected in a 50 mL beaker and evaporated to a final volume of one mL. The final volume was combined with 100 mL clean dilution water to create the eluate sample for toxicity testing with *H. azteca*. An Ambersorb elution blank was prepared by performing the above treatments on Ambersorb that had been combined with formulated sediment. A 1% acetone blank was also tested.

• SIR-300 (ResinTech, West Berlin, NJ) is a macroporous weak acid cation exchange resin based on the iminodiacetate acid functional group, which has chelating properties for heavy metal ions even in conditions with high calcium concentrations. After preparation, SIR-300 can be mixed into sediment to reduce cationic metal bioavailability (Burgess et al. 2000). Ten percent SIR-300 (wet weight) was added to the sediment in a 500 mL mixing jar. Treated sediment was homogenized for 24 hours on a roller apparatus, and loaded into exposure chambers. A dilution blank was created by combining test sediment with 10% formulated sediment, and an SIR-300 blank was created by adding 10% SIR-300 to formulated sediment. At test termination the sediment was sieved through a series of screens ranging from 250-400 μm to retain the SIR-300. The SIR-300 was then eluted by loading a column with approximately 7.5g resin and pumping 10 mL of 1N hydrochloric acid through the column at a rate of 1 mL per minute. Post-column acid was combined with 100 mL clean dilution water and neutralized to create the eluate sample for toxicity testing with *H. azteca*. An SIR-300 elution blank was prepared by performing the above treatments on SIR-300 that had been combined with formulated sediment. An acid blank was also tested.

• The enzyme carboxylesterase (Sigma-Aldrich, St. Louis, MO) hydrolyzes ester-containing compounds such as pyrethroids to their corresponding acid and alcohol, which are generally not toxic (Wheelock et al. 2004). Carboxylesterase (500x) was added to the overlying water on the day of test initiation, six hours prior to the addition of amphipods. This allowed for interaction between the enzyme and pyrethroids. The enzyme was added based on units of activity. One 'x' of enzyme activity equals 0.0025 units of enzyme per mL of sample, therefore at 500x, 1.25 units per mL were added. Enzyme strength is unique for each lot purchased (Wheelock et al. 2004). To control for the binding of contaminants to the protein base of the enzyme, a separate set of replicates was treated with bovine serum albumin (BSA). Reduction of toxicity by the enzyme, and not the BSA, would confirm the presence of pyrethroids. The enzyme and protein treatments were given daily booster shots of BSA and carboxylesterase.

• Piperonyl butoxide (Sigma-Aldrich, St. Louis, MO) is used to block the metabolic activation of acetylcholinesterase-inhibiting organophosphate pesticides (Ankley et al. 1995). It is also a potent synergist of pesticide toxicity, because it inhibits their metabolism (Ware 1989, Kakko et al. 2000). The PBO treatment contained 500 μg/L of PBO in the water overlying the sediment. Decreased toxicity with the addition of PBO suggests the presence of organophosphate pesticides. Increased toxicity with the addition of PBO suggests the presence of pyrethroids. There is also some evidence that PBO increases toxicity caused by DDT (Brandt et al. 2002), but we are unaware of specific studies exposing *H. azteca* to mixtures of PBO and DDT.

• An additional baseline treatment was conducted at 15ºC because the toxicity of some organochlorine and pyrethroid pesticides increases with decreasing temperature (Ware 1989).

Interstitial Water Treatments

The following Phase 1 TIE treatments were performed on a dilution series of each sample (US EPA 1991). Sample concentrations in the initial test were 0 (treatment blank), 10, 25, 50, and 100%. The treatment blank was control water that underwent the same manipulation as the sample.

• Baseline - Toxicity test on un-manipulated sample. Concentrations were chosen to bracket the effect concentration of the sample and might differ from initial test.

• Cation Column - The Cation Column is designed to remove metals from the sample. The column was eluted with 1N hydrochloric acid (HCl) and the resulting eluate was tested to determine if substances removed by the column were toxic.

• HLB Column - The HLB Column is designed to remove non-polar organic compounds from the sample. In the manipulation, reverse phase liquid chromatography was applied to extract nonionic organic toxicants from the aqueous sample. The column was eluted with methanol and the resulting eluate was tested to determine if substances removed by the column were toxic.

Sequential HLB Cation Column – The two solid-phase extraction columns are used in sequence to determine if toxicity was caused by both metals and organics. Each column is individually eluted to determine if substances removed by the columns were toxic.

• Carboxylesterase – As in the solid-phase treatment, carboxylesterase was added to the sample to break down suspected pyrethroid pesticides (Wheelock et al. 2004). BSA was added in a separate treatment to control for the binding of contaminants to the protein base of the enzyme

• PBO was added to the interstitial water to determine if organophosphate or pyrethroid pesticides were causing toxicity.

Interstitial water exposures were conducted in 20 mL glass scintillation vials (3 replicates) containing 15 mL treated sample and five amphipods. Acute exposures were conducted for 96 hours, following US EPA 1993.

Physical and Chemical Measurements

Water quality parameters of dissolved oxygen, pH and conductivity were measured using a Hach SensION© selective ion meter with appropriate electrodes; and ammonia was measured using a Hach 2010 spectrophotometer. Temperature was measured using a continuously recording thermograph and thermometer. Concentrations of the organophosphate pesticides chlorpyrifos and diazinon were measured using enzyme-linked immunosorbent assays (ELISA, Strategic Diagnostics Inc, Newark, DE).

Data Interpretation

Treatment blanks were evaluated to determine if sample manipulations added toxic artifacts. Treatment data were then compared to one another using the toxic unit approach or simply based on organism response. Toxic units (TU) were calculated by dividing 100 by the LC50 calculated from each treatment dilution series. A lower toxic unit value indicates a treatment has been effective in reducing toxicity.

Results and Discussion

Initial Tests

Survival in both initial sediment tests was 0% (Table 1). Interstitial water from DPCCR sediment caused complete mortality event when diluted to 10% strength (20 toxic units). WWNCR interstitial water was less toxic at 2.2 TU. Interstitial water was also analyzed for chlorpyrifos and diazinon using enzyme-linked immunosorbent assays (ELISA). The concentrations of diazinon and chlorpyrifos were below reporting limits. Interstitial water and solid-phase TIEs were pursued for both samples. Water quality parameters for all TIEs were within acceptable limits for the test organism (Tables 2-5)

DPCCR TIEs

Addition of enzyme to the overlying water was the only treatment that increased survival in the solid-phase TIE for DPCCR. Addition of enzyme increased survival from 0% to 22% (Table 6), whereas the addition of BSA did not increase survival. These results indicate the cause of toxicity was an organic contaminant and possibly a pyrethroid pesticide. Decreasing test temperature did not increase toxicity because complete mortality was observed in the standard baseline treatment (data not shown).

Although the addition of Ambersorb to the sediment did not reduce toxicity, the resin was isolated from the test sediment at the termination of the exposure and eluted with acetone. When the acetone was added back to clean dilution water it caused significant toxicity indicating an organic was removed from the sediment by the Ambersorb and was successfully returned to clean dilution water (Table 6).

Based on the toxicity results, the sediment and acetone extract were analyzed for organochlorine, organophosphate, and pyrethroid pesticides. The sediment contained low concentrations of DDE (p,p') and elevated concentrations of several pyrethroids (Table 10). The sediment contained 11.9 toxic units of bifenthrin after organic carbon correction (Amweg et al. 2004). Although the acetone extract of the Ambersorb was significantly toxic, there were no pyrethroids detected in the solvent. The Ambersorb extract only contained a small concentration of DDE (p,p') (data not shown).

The DPCCR interstitial water TIE produced similar results to the solid-phase TIE (Table 7). The enzyme was the most effective treatment at reducing toxicity. Addition of the enzyme to interstitial water reduced toxicity from 18.6 TU to <1 TU. Addition of BSA did not significantly alter the toxicity indicating that

pyrethroid pesticides are the most likely cause of toxicity. The HLB column did not significantly reduce toxicity until combined with the cation column. Both
HLB eluates returned toxicity to clean dilution water, indicating the presence of organic contaminants. PBO did not appear to increase toxicity because all dilutions of the interstitial water caused complete mortality.

Interstitial water was not analyzed for pesticides because of insufficient volume, but the HLB solvent eluates were analyzed for organochlorine, organophosphate, and pyrethroid pesticides. No pesticides were detected in the column extracts.

WWNCR TIEs

The results of the WWNCR TIEs were similar to those of DPCCR. Addition of enzyme to the overlying water was the only treatment that increased survival in the solid-phase TIE for WWNCR. Addition of enzyme increased survival from 0% to 48% (Table 8), and the addition of BSA only increased the survival to 8%. These results indicate the cause of toxicity was an organic contaminant and possibly a pyrethroid pesticide. Decreasing test temperature did not increase toxicity because complete mortality was observed in the standard baseline treatment (data not shown).

The addition of Ambersorb to WWNCR sediment did not reduce toxicity, but the resin was isolated at the termination of the exposure and eluted with acetone. When the acetone was added back to clean dilution water it caused significant toxicity indicating an organic was removed from the sediment by the Ambersorb and was successfully returned to clean dilution water (Table 9).

The sediment contained low concentrations of DDE (p,p') and elevated concentrations of several pyrethroids (Table 10). The sediment contained just over one TU of bifenthrin and 31.1 TUs of lambda-cyhalothrin (organic carbon corrected concentrations, Amweg et al. 2004). Although the acetone extract of the Ambersorb was significantly toxic, there were no pyrethroids detected in the solvent. The Ambersorb extract only contained a small concentration of DDE (p,p') (data not shown).

Toxicity of the WWNCR interstitial water was reduced by the column treatments and the enzyme (Table 9). The cation column reduced toxicity from 5.6 TU to 1.5 TU, but there was no toxicity in the cation eluates, indicating that metals were unlikely to be the cause of toxicity. The HLB column reduced toxicity to 1.9 TUs, but returned 6.5 TUs, indicating the cause of toxicity was an organic contaminant. The enzyme reduced toxicity to 1.7 TUs and the BSA slightly reduced toxicity to 4.4 TUs. These results confirm the characterization of an organic contaminant and indicate the cause of toxicity was a pyrethroid. Addition of PBO increased toxicity to 18.5 TUs providing further evidence that the cause of toxicity was a pyrethroid.

Interstitial water was not analyzed for pesticides because of insufficient volume, but the HLB solvent eluates were analyzed for organochlorine, organophosphate, and pyrethroid pesticides. No pesticides were detected in the column extracts.

Conclusions

Implicating the class of chemicals responsible for the cause of sediment toxicity requires multiple lines of evidence that include the results of TIE treatments, as well as chemical analyses. The enzyme reduced toxicity in both solid-phase TIEs, while the BSA did not significantly alter the responses. The Ambersorb did not reduce toxicity, a significant signal was returned to clean dilution water in both cases. The enzyme also reduced toxicity in both interstitial water TIEs with no reduction in the BSA treatments. The PBO treatment in the WWNCR interstitial water TIE increased toxicity three times. DPCCR sediment contained 11.9 TUs of bifenthrin and WWNCR sediment contained 31.1 TUs of lambda-cyhalothrin. Although toxicity was returned in the eluates of the Ambersorb and the HLB columns, no significant chemical concentrations were detected.

The solid-phase and interstitial water Phase I TIE treatments characterized the cause of toxicity as an organic. Additional Phase II treatments (enzyme and BSA) and solid-phase chemical analysis identified the causes of toxicity as pyrethroid pesticides. Because pyrethroids were not detected in the solvent eluates of the Ambersorb and HLB treatments, we were unable to confirm the return of pyrethroids to clean dilution water.

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Table 1. Mean percent survival and standard deviation (SD) of *H. azteca* in initial solid-phase and interstitial water tests.

	DPCCR				WWNCR			
	Solid-phase		Interstitial		Solid-phase		Interstitial	
Concentration	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Control	96	5	100	0	96	5	100	0
10%			0	0			100	0
25%			0	0			87	12
50%			0	0			53	12
100%	0	0	0	0	0	0	0	0

Table 2. Water quality measurements for the DPCCR solid-phase TIE. ND indicates non-detect. NA indicates not analyzed.

	pH		Dissolved Oxygen (mg/L)		Conductivity (µS/cm)		Total Ammonia (mg/L)		Un-ionized Ammonia (mg/L)	
Treatment	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Baseline DPCCR	8.30	8.41	8.14	8.69	688	703	ND	ND	ND	ND
DPCCR (10% Ambersorb)	8.43	8.42	8.21	8.60	705	728	0.2	ND	0.023	ND
Control (10% Ambersorb)	8.50	8.45	8.16	8.56	880	881	0.2	ND	0.027	ND
DPCCR (10% SIR-300)	8.60	8.65	8.15	8.45	876	889	0	0.2	0.000	0.036
Control (10% SIR-300)	8.58	8.85	7.93	8.54	1076	1213	0.6	0.2	0.095	0.052
DPCCR (10% Control)	8.43	8.49	8.22	8.59	800	769	ND	ND	ND	ND
DPCCR (Enzyme)	8.35	8.28	7.99	7.43	659	793	0.9	19.0	0.089	1.632
Control (Enzyme)	8.38	8.32	8.08	6.87	862	974	0.6	24.2	0.063	2.261
DPCCR (BSA)	8.38	8.29	8.21	7.39	750	802	ND	14.0	ND	1.228
Control (BSA)	8.45	8.28	7.95	6.49	880	985	0.8	25.2	0.098	2.165
DPCCR (PBO)	8.47	8.46	8.20	8.30	797	707	0.3	0	0.038	0.000
Control (PBO)	8.03	8.54	8.13	8.59	790	891	0.3	0.3	0.015	0.044
Control	8.39	8.54	8.13	8.62	739	903	0.4	0.2	0.043	0.029

Table 3. Water quality measurements for the DPCCR interstitial water TIE. ND indicates non-detect. NA indicates not analyzed.

	pH		Dissolved Oxygen (mg/L)		Conductivity (µS/cm)		Total Ammonia (mg/L)		Un-ionized Ammonia (mg/L)	
Treatment	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Baseline	8.21	NA	8.24	NA	921	NA	1.5	NA	0.111	NA
Cation Column	8.32	NA	8.35	NA	898	NA	1.2	NA	0.112	NA
Cation Eluate	8.10	NA	8.28	NA	5850	NA	NA	NA	NA	NA
HLB Column	8.32	NA	8.33	NA	922	NA	4.5	NA	0.420	NA
HLB Eluate	8.47	NA	8.34	NA	868	NA	NA	NA	NA	NA
Sequential Cation HLB	8.38	NA	8.28	NA	900	NA	1.3	NA	0.138	NA
Sequential Cation Eluate	7.89	NA	8.31	NA	5640	NA	NA	NA	NA	NA
Sequential HLB Eluate	8.49	NA	8.33	NA	643	NA	NA	NA	NA	NA
Enzyme	8.18	NA	8.27	NA	969	NA	2.0	NA	0.139	NA
BSA	8.18	NA	8.21	NA	925	NA	1.6	NA	0.111	NA
PBO	8.24	NA	8.23	NA	915	NA	1.3	NA	0.103	NA

Table 4. Water quality measurements for the WWNCR solid-phase TIE. ND indicates non-detect. NA indicates not analyzed.

	pH		Dissolved Oxygen (mg/L)		Conductivity (µS/cm)		Total Ammonia (mg/L)		Un-ionized Ammonia (mg/L)	
Treatment	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Baseline WWNCR	8.50	8.62	8.02	8.02	919	940	4.4	0.3	0.594	0.051
WWNCR (10% Ambersorb)	8.45	8.55	8.00	8.47	793	831	4.9	ND	0.598	ND
Control (10% Ambersorb)	8.50	8.45	8.16	8.56	880	881	0.2	ND	0.027	ND
WWNCR (10% SIR-300)	8.41	8.76	7.35	8.45	1008	1030	0.6	0.2	0.068	0.044
Control (10% SIR-300)	8.58	8.85	7.93	8.54	1076	1213	0.6	0.2	0.095	0.052
WWNCR (10% Control)	8.25	8.56	7.27	8.44	785	820	3.2	ND	0.258	ND
WWNCR (Enzyme)	8.18	8.15	7.02	7.63	826	843	4.7	2.1	0.326	0.137
Control (Enzyme)	8.38	8.32	8.08	6.87	862	974	0.6	24.2	0.063	2.261
WWNCR (BSA)	8.44	8.27	1.09	7.43	954	904	4.1	4.7	0.490	0.395
Control (BSA)	8.45	8.28	7.95	6.49	880	985	0.8	25.2	0.098	2.165
WWNCR (PBO)	8.30	8.68	3.13	8.39	931	896	4.2	ND	0.376	ND
Control (PBO)	8.03	8.54	8.13	8.59	790	891	0.3	0.3	0.015	0.044
Control	8.39	8.54	8.13	8.62	739	903	0.4	0.2	0.043	0.029

Table 5. Water quality measurements for the WWNCR interstitial water TIE. ND indicates non-detect. NA indicates not analyzed.

	pH		Dissolved Oxygen (mg/L)		Conductivity (μS/cm)		Total Ammonia (mg/L)		Un-ionized Ammonia (mg/L)	
Treatment	Initial	Final	Initial	Final	Initial	Final	Initial	Final	Initial	Final
Baseline	7.39	NA	1.38	NA	3000	NA	26.0	NA	0.311	NA
Cation Column	7.76	NA	2.48	NA	2910	NA	24.2	NA	0.668	NA
Cation Eluate	8.13	NA	8.52	NA	4760	NA	NA	NA	NA	NA
HLB Column	7.81	NA	4.34	NA	2910	NA	24.4	NA	0.753	NA
HLB Eluate	8.20	NA	8.91	NA	742	NA	NA	NA	NA	NA
Sequential Cation HLB	7.89	NA	4.48	NA	2890	NA	24.4	NA	0.900	NA
Sequential Cation Eluate	7.94	NA	8.85	NA	4710	NA	NA	NA	NA	NA
Sequential HLB Eluate	8.34	NA	8.75	NA	710	NA	NA	NA	NA	NA
Enzyme	7.40	NA	0.57	NA	2960	NA	21.0	NA	0.257	NA
BSA	7.41	NA	0.49	NA	2970	NA	24.7	NA	0.309	NA
PBO	7.43	NA	0.70	NA	2980	NA	25.5	NA	0.334	NA

Table 6. Mean percent survival and standard deviation (SD) of *H. azteca* in solid-phase DPCCR TIE.

Treatment	Solid-Phase Treatments Mean	Solid-Phase Treatments SD	Acetone Eluate Treatments Mean	Acetone Eluate Treatments SD
Baseline DPCCR	0	0		
DPCCR (10% Ambersorb)	0	0	0	0
Control (10% Ambersorb)	98	4	88	11
DPCCR (10% SIR-300)	0	0		
Control (10% SIR-300)	94	5		
DPCCR (10% Control)	0	0		
DPCCR (Enzyme)	22	22		
Control (Enzyme)	100	0		
DPCCR (BSA)	0	0		
Control (BSA)	82	8		
DPCCR (PBO)	0	0		
Control (PBO)	96	9		
Elution Control			81	2
Control	94	9	93	12

Table 7. Mean percent survival and standard deviation (SD) of *H. azteca* in interstitial water DPCCR TIE.

	Percent Sample										
	0%		10%		25%		50%		100%		Toxic
Treatment	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Units
Baseline	93	12	7	12	0	0	0	0	0	0	18.6
Cation Column	90	9	0	0	0	0	0	0	0	0	20.0
Cation Eluate	81	2	62	20	80	0	93	12	87	12	<1
HLB Column	93	12	0	0	0	0	0	0	0	0	20.0
HLB Eluate	93	12	89	19	20	35	0	0	0	0	5.6
Sequential Cation HLB	80	0	33	12	0	0	0	0	0	0	11.7
Sequential Cation Eluate	80	0	73	12	73	12	80	0	100	0	<1
Sequential HLB Eluate	93	12	100	0	87	12	47	50	0	0	2.2
Enzyme	100	0	100	0	100	0	100	0	93	12	<1
BSA	100	0	0	0	0	0	0	0	0	0	20.0
PBO	88	11	0	0	0	0	0	0	0	0	20.0

Table 8. Mean percent survival and standard deviation (SD) of *H. azteca* in solid-phase WWNCR TIE.

	Solid-Phase Treatments		Acetone Eluate Treatments	
Treatment	Mean	SD	Mean	SD
Baseline WWNCR	0	0		
WWNCR (10% Ambersorb)	0	0	0	0
Control (10% Ambersorb)	98	4	88	11
WWNCR (10% SIR-300)	0	0		
Control (10% SIR-300)	94	5		
WWNCR (10% Control)	0	0		
WWNCR (Enzyme)	48	37		
Control (Enzyme)	100	0		
WWNCR (BSA)	8	11		
Control (BSA)	82	8		
WWNCR (PBO)	0	0		
Control (PBO)	96	9		
Elution Control			81	2
Control	94	9	93	12

Table 9. Mean percent survival and standard deviation (SD) of *H. azteca* in interstitial water WWNCR TIE.

	Percent Sample										
	0%		10%		25%		50%		100%		Toxic
Treatment	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Units
Baseline	76	8	100	0	0	0	13	12	0	0	5.6
Cation Column	100	0	93	12	100	0	93	12	0	0	1.5
Cation Eluate	93	12	100	0	100	0	100	0	93	12	<1
HLB Column	93	12	100	0	67	23	87	12	0	0	1.9
HLB Eluate	93	12	88	11	0	0	0	0	0	0	6.5
Sequential Cation HLB	100	0	100	0	73	31	73	12	0	0	2.1
Sequential Cation Eluate	93	12	47	42	100	0	100	0	93	12	<1
Sequential HLB Eluate	93	12	100	0	80	20	100	0	27	12	1.3
Enzyme	100	0	100	0	87	12	87	23	0	0	1.7
BSA	93	12	100	0	33	23	13	23	0	0	4.4
PBO	93	12	7	12	0	0	0	0	0	0	18.5

Table 10. Concentrations of detected pesticides in DPCCR and WWNCR sediment. Sediment LC50 values. ND indicates not detected. NR indicates not reported. *Amweg et al. (2004).

Chemical	DPCCR ng/g dry wt.	WWNCR ng/g dry wt.	Sediment LC50* ng/g dry wt.
DDE (p,p')	58.9	67.1	
Bifenthrin	58.6	5.08	12.9
Bifenthrin µg/g oc	6.17	0.63	0.52
(Es)Fenvalerate	1.09	ND	41.8
(Es)Fenvalerate µg/g oc	0.11	ND	1.54
Lambda-cyhalothrin	2.56	113.7	5.6
Lambda-cyhalothrin µg/g oc	0.27	14.0	0.45
Permethrin	4.92	ND	201
Permethrin µg/g oc	0.52	ND	10.8
Total Organic Carbon	0.95%	0.81%	